Analysis of Persistent Organic Pollutants in Environmental Samples: Analytical Challenges

Eric J. Reiner

**Laboratory Services Branch,
Ministry of the Environment, 125 Resources Road,
Toronto, ON, M9P 3V6**

Ministry of the
Environment

ONTARIO

Persistent Organic Pollutants - Outline

- Analytical Challenges – POPs,Dioxins and Dioxin-like compounds
- Emergency Response / Reduced Analysis times
- Fast GC / Dual Column analysis
- Other POPs (PCNs, BFRs)
- Future work

What is a Persistent Organic Pollutant?

UNEP (United Nations Environmental Programme) definition:

- Chemical Substances that persist in the Environment
- Bioaccumulate through the food web
- Pose a risk of causing adverse affects to human health
- Evidence of long range transport to regions where they have never been used

What is a Persistent Organic Pollutant?

Classical POPs – The Dirty Dozen:

Aldrin, DDT (and metabolites), Dieldrin, Endrin, Chlordane (technical), Heptachlor, Hexachlorobenzene, Mirex, Toxaphene, Polychlorinated Dioxins and Furans, Polychlorinated Biphenyls (PCBs)

New POPs:

Brominated Flame Retardants, Polychlorinated Naphthalenes, Dioxin-Like PCBs, Polychlorinated Diphenylethers

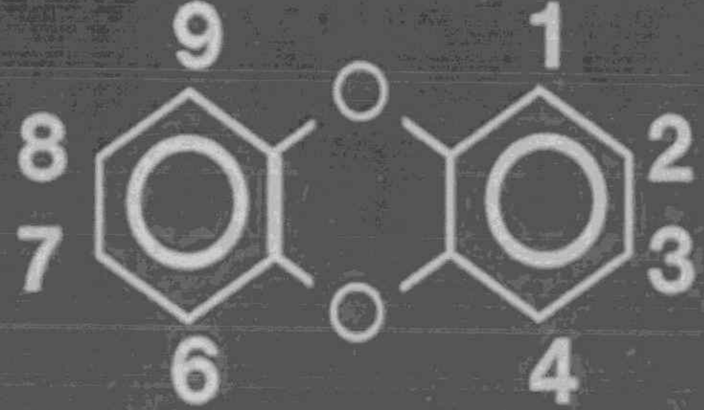
Chemical	LD_{50} Oral - Rat[2] (mg/Kg)
Hexachlorobenzene	10,000
[illegible]-BHC	6,000
Pyrene	2,700
PCBs - Aroclor	1,000
DDE	880
2,4-D	370
2,4,5-T	300
Mirex	235
Chlordane	200
DDT	87
Lindane	76
Toxaphene	50
Benzo(a)pyrene	50 (scu)
Aldrin	39
Dieldrin	38
Pentachorophenol	27
Endosulfan	18
Endrin	3
PCB 126	0.2
2,3,7,8-TCDD	0.02

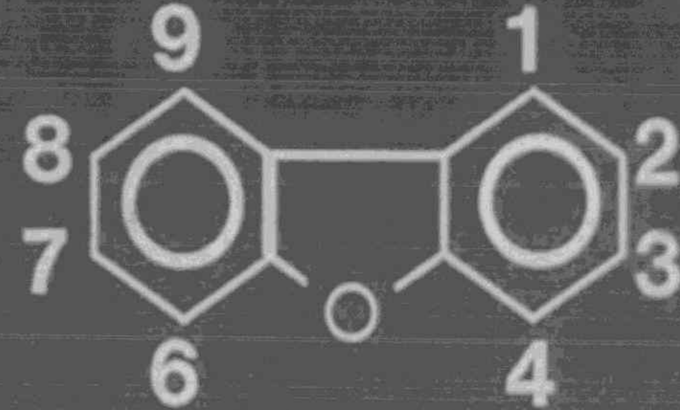
POPs in Environmental Samples: The Analytical Challenge

- **Many congeners per analyte group**
  [dioxins/furans: 210; PCBs: 209, Toxaphene: >600]
- **Separate and accurately quantify all toxic congeners**
  [dioxins/furans: 17, PCBs: 12, Toxaphene: 22]
- **Toxicity can range up to 6 orders of magnitude**
  [NOEL = 3g/kg to LD_{50} = 1 ug/kg]
- **Range of concentrations** [fg/g (10^{-15}g/g) to %]
- **Range of sample types, complexities**
  [biota, air, water, soil, haz. waste, other]

Compound Class	Detection Range	Analytical Method	Sample Size	Sample Cleanup	Analysis Time	Extract Volume	Cost ($)
Chlorobenzene	ug/g - ng/g	GC-ECD	1 g	Florisil or Silica	1 to 2 days	5 mL	$50 to $100
Polychlorinated Biphenyls (total)	ug/g - ng/g	GC-ECD	1 g	Florisil	1 to 2 days	2 to 5 mL	$50 to $150
Organochlorine Pesticides	ng/g	GC-ECD	1 g	Florisil	2 days	2 to 5 mL	$100 to $200
PCBs -Congener	ng/g - pg/g	GC-ECD GC/MS	1 - 5 g	Florisil	2 to 3 days	0.5 to 5 mL	$100 to $250
Polyaromatic Hydrocarbons	ng/g	GC/MS	1 - 5 g	Silica	3 days	0.1 to 0.5 mL	$100 to $200
Brominated Diphenylethers	low ng/g	GC/MS GC/HRMS	1 - 5g	Silica	3 to 4 days	0.1 to 0.5 mL	$400 to $600
Polychlorinated Naphthalenes	low ng/g	GC/ECD GC/MS-NCI	1 - 5g	Silica/ Carbon	2 to 3 days	0.1 to 0.5 mL	$400 to $600
PCBs (Dioxin - like)	low pg/g	GC/MS GC/HRMS	5 - 10 g	Silica, Carbon	5 to 6 days	10 to 100 uL	$400 to $600
Dioxins/Furans	pg/g - fg/g	GC/HRMS GC/MS/MS	5 - 20 g	Silica/ Alumina/ Carbon	6 to 8 days	10 to 20 uL	$ 800 to $1000

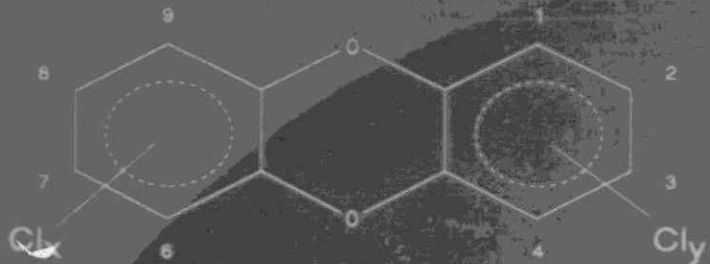
Dibenzo-p-dioxin   Dibenzofuran

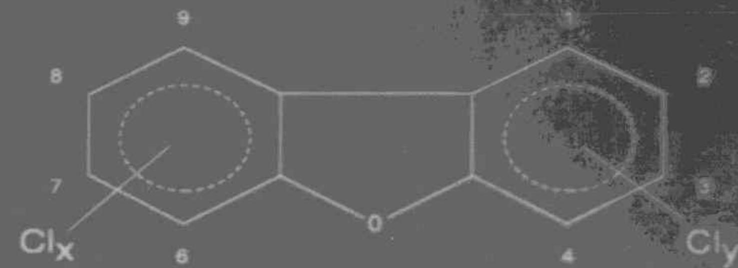




number of chlorines	number of isomers PCDD	number of isomers PCDF
1	2	4
2	10	16
3	14	28
4	22	38
5	14	28
6	10	16
7	2	4
8	1	1
total	75	135

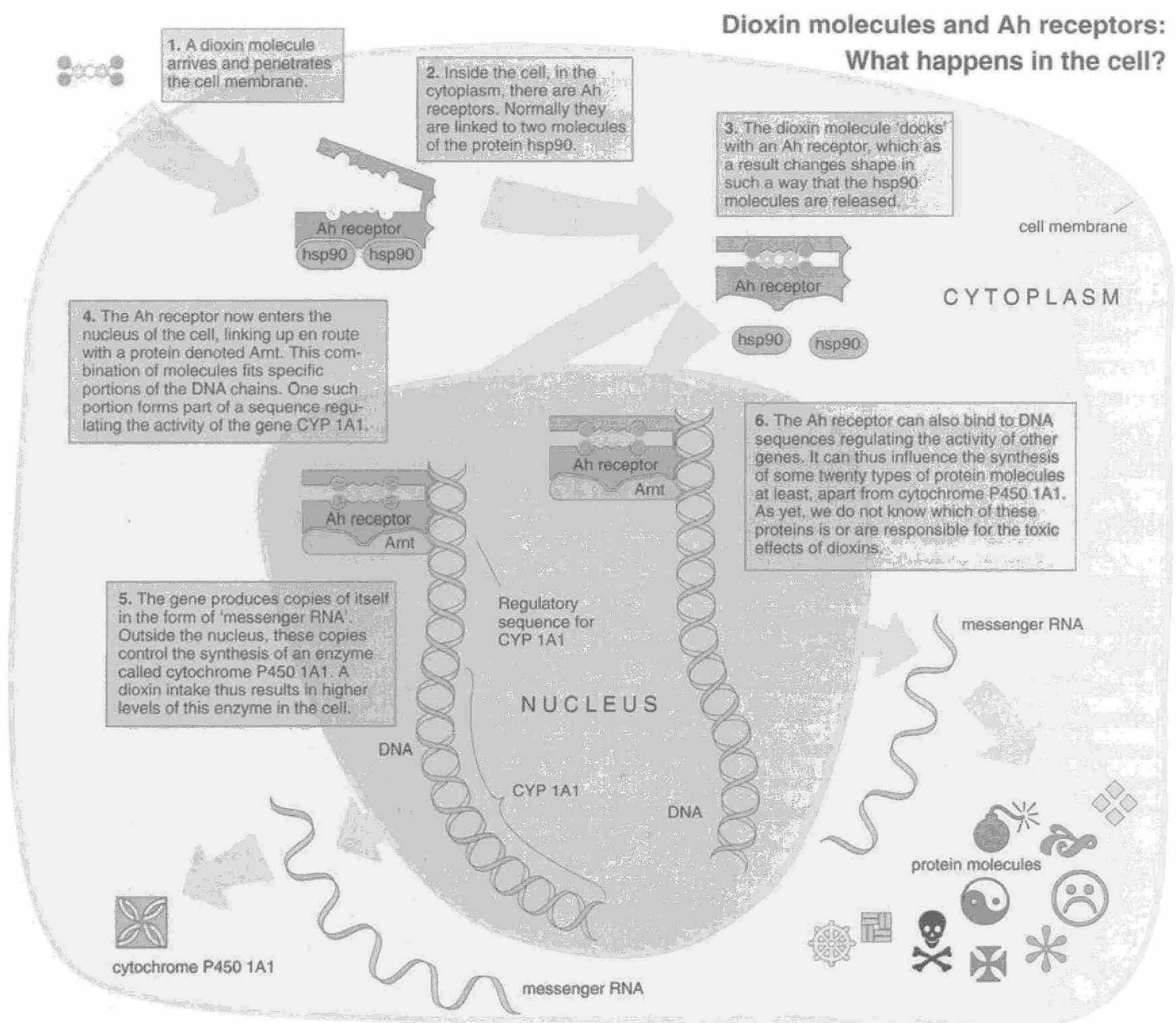
International Toxicity Equivalency Factors





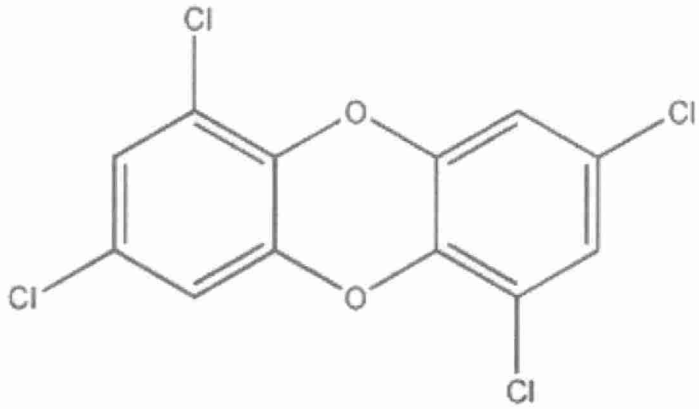
2,3,7,8-TCDD	1	2,3,7,8-TCDF	0.1
1,2,3,7,8-P5CDD	1	2,3,4,7,8-P5CDF	0.5
		1,2,3,7,8-P5CDF	0.05
1,2,3,4,7,8-H6CDD	0.1	1,2,3,4,7,8-H6CDF	0.1
1,2,3,7,8,9-H6CDD	0.1	1,2,3,7,8,9-H6CDF	0.1
1,2,3,6,7,8-H6CDD	0.1	1,2,3,6,7,8-H6CDF	0.1
		2,3,4,6,7,8-H6CDF	0.1
1,2,3,4,6,7,8-H7CDD	0.01	1,2,3,4,6,7,8-H7CDF	0.01
		1,2,3,4,7,8,9-H7CDF	0.01
OCDD	0.0001	OCDF	0.0001

WHO (1997)

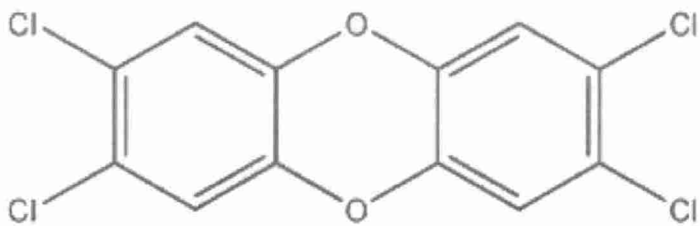


After Wærn 1995.

Dioxin Toxicity



1,3,6,8 Tetrachlordibenzo-p-dioxin

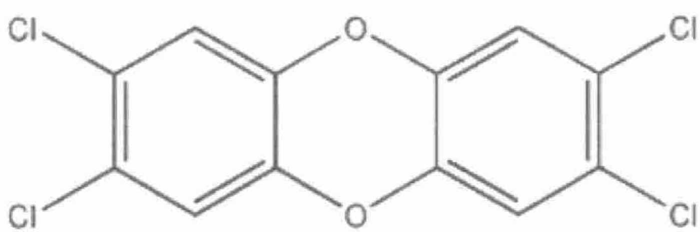


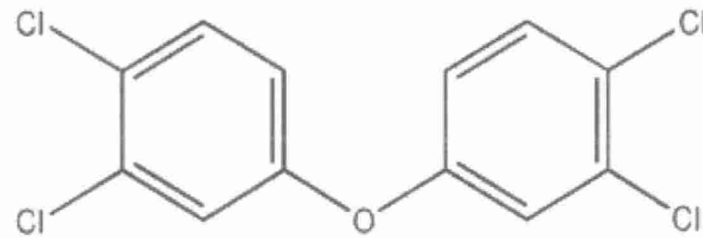
2,3,7,8 Tetrachlordibenzo-p-dioxin

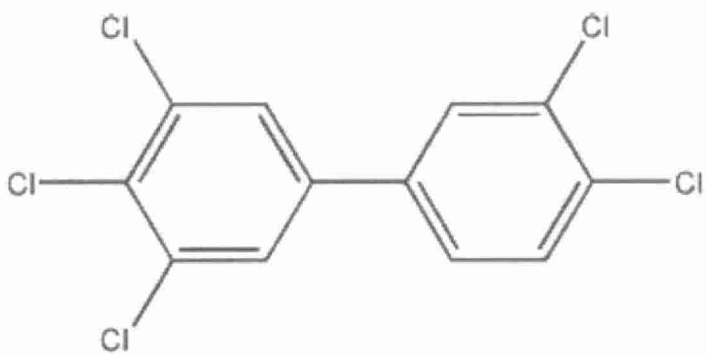
NOEL = 3g / kg

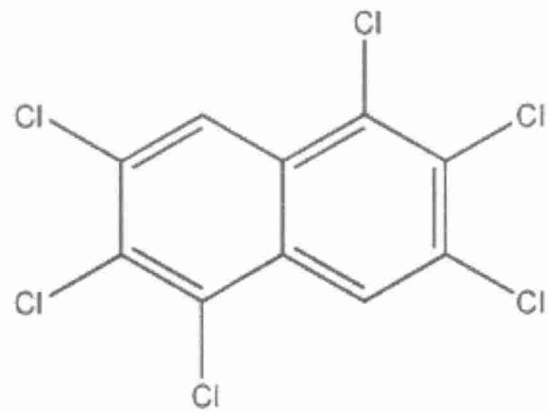
LD_{50} = 1ug / kg

Dioxin-like Toxicity


2,3,7,8 Tetrachlordibenzo-p-dioxin


3,3',4,4' Tetrachlorodiphenylether


3,3',4,4',5 Pentachlorobiphenyl


1,2,3,5,6,7 Hexachloronaphthalene

Most toxic are planar with 3Å x 10Å dimensions

Compounds With Dioxin-Like Activity

TEF	COMPOUND
1	2,3,7,8-TCDD 1,2,3,7,8-P_5CDD
0.5	2,3,4,7,8-P_5CDF
0.1	2,3,7,8-TCDF, 2,3,7,8-Substituted H_6CDD/Fs, PCB-126
0.05	1,2,3,7,8-P_5CDF
0.01	2,3,7,8-Substituted H_7CDD/Fs, PCB-169
0.001-0.003	Coplanar PCDEs, H_6CNs
0.0002	Mono-ortho PCDEs
0.0001	OCDD/F, PCBs, - 77, 81, 105, 118, 123, 189
0.0005	PCB-114, PCB-156, PCB-157
0.00001	PCB-167

POPs in Environmental Samples: The Analytical Challenge

- Quantitatively extract analytes from matrix
- Extract may contain up to 1 gram of co-extractable organic material (PAH, PCB, OC, BFR, PCN, PCDE)
- Extract must be cleaned to remove interfering co-extractables
- Separate target analytes from non-target isomers or congeners
- Detect analytes at sub picogram level – every piece of labware must be prechecked to contain less than 0.5 pg
- Ensure instrument selectivity and sensitivity to meet Data Quality Objectives (DQOs)
- Ensure quantitative accuracy

POPs in Environmental Samples: The Analytical Challenge

- Complex multistage column cleanup required to remove co-extractable compounds
- Complex Gas Chromatography / High Resolution Mass Spectrometry (GC/HRMS) using isotope dilution
- Highly skilled analysts
- Ultra trace detection limits (100 fg (100×10^{-15}g))
- Numerous hazards – toxic standards/chemicals and samples, high voltages, sharp objects

Advantages of Isotope Dilution MS

- Results are calculated using a ratio of the native analyte corrected for concentration to the $^{13}C_{12}$ labeled dioxin / furan surrogate signals (areas from chromatogram)
- The slope of this plot is the response factor and is typically 1 $\pm$ 0.1
- Concentration is:

$$C = A_n / A_l \times rf \times v/m$$

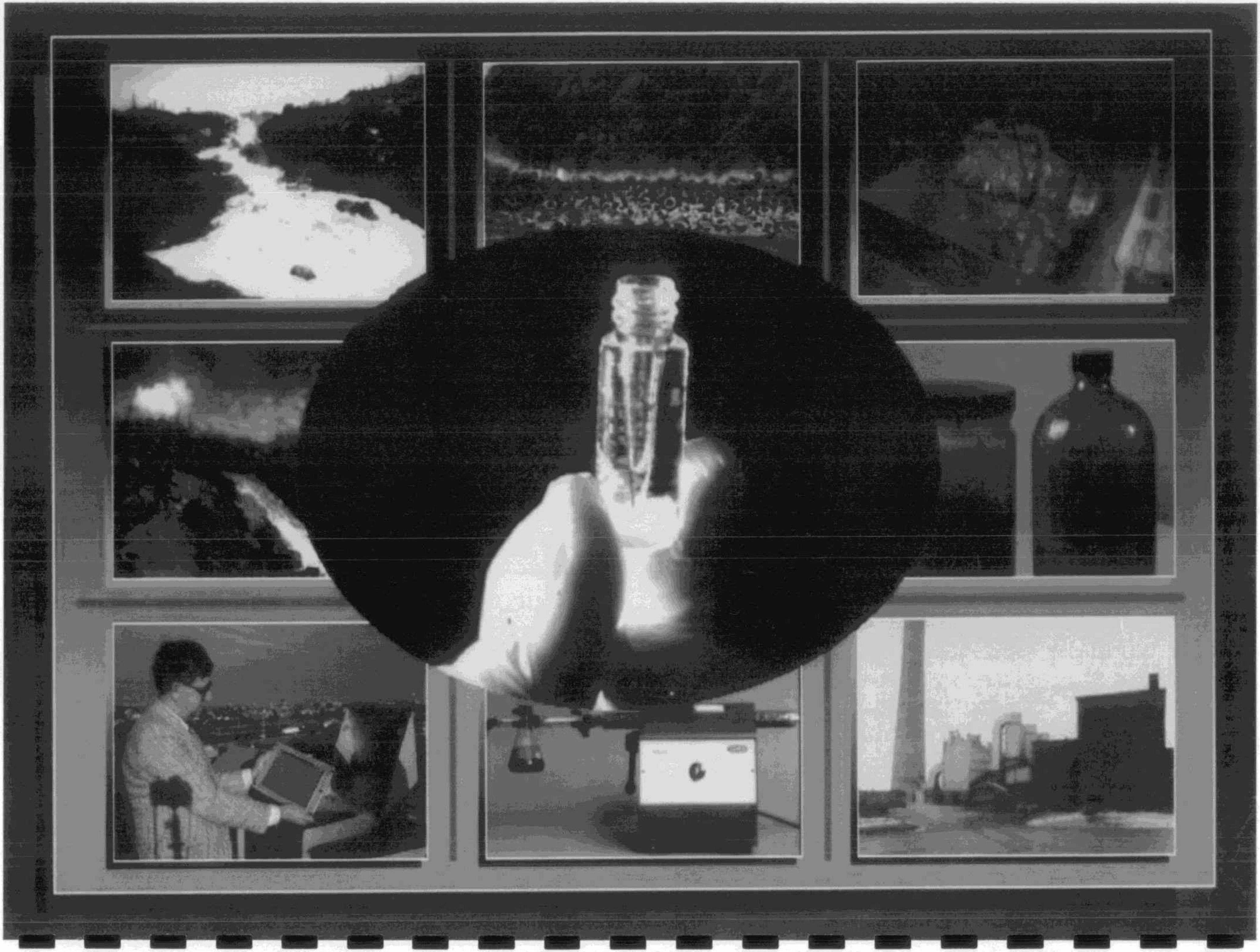
Where A_n = area of native

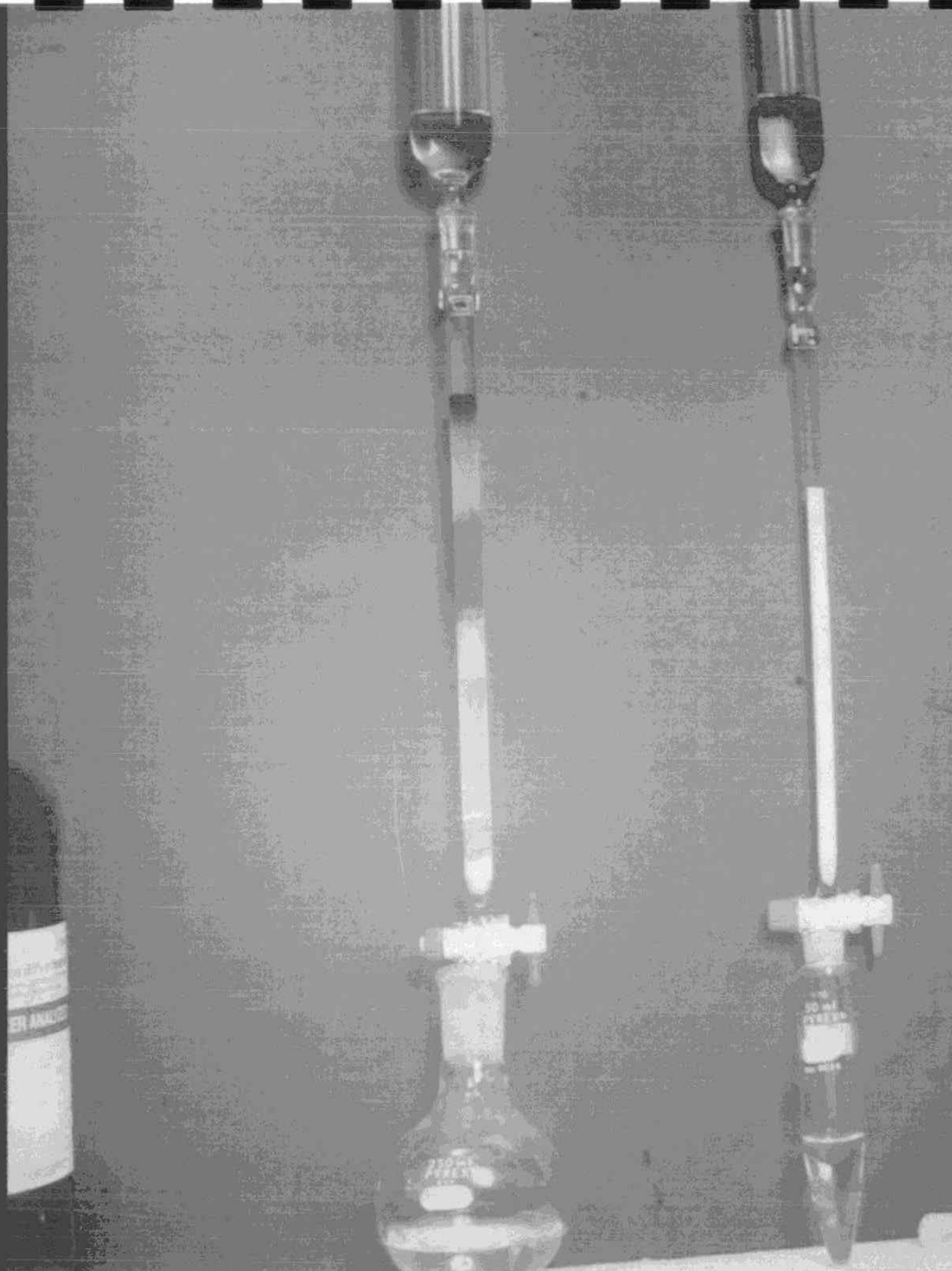
A_l = area of surrogate

rf = response factor

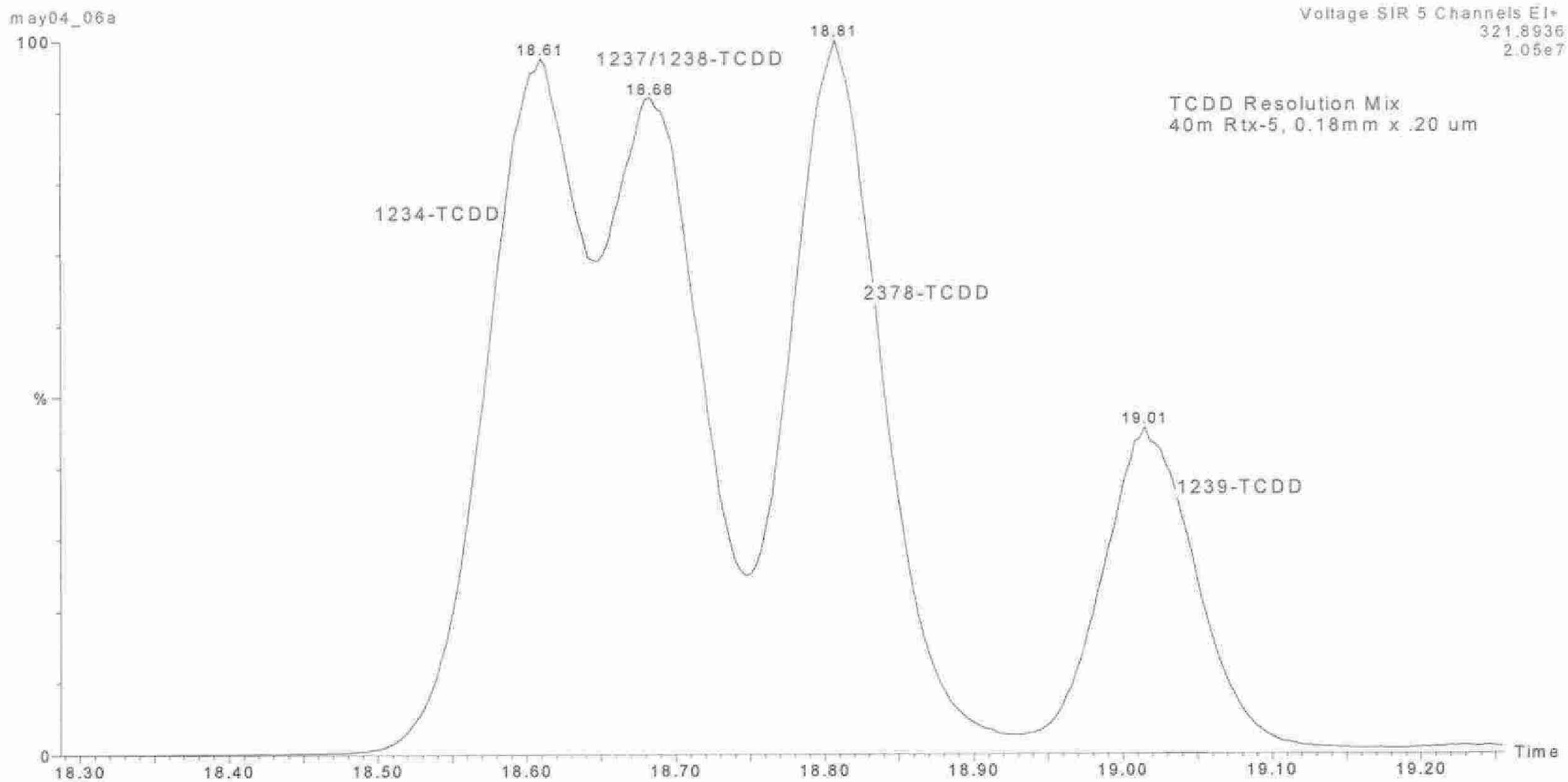
v/m = volume or mass

- Surrogate standard acts as a carrier
- Results are automatically corrected for loss during processing





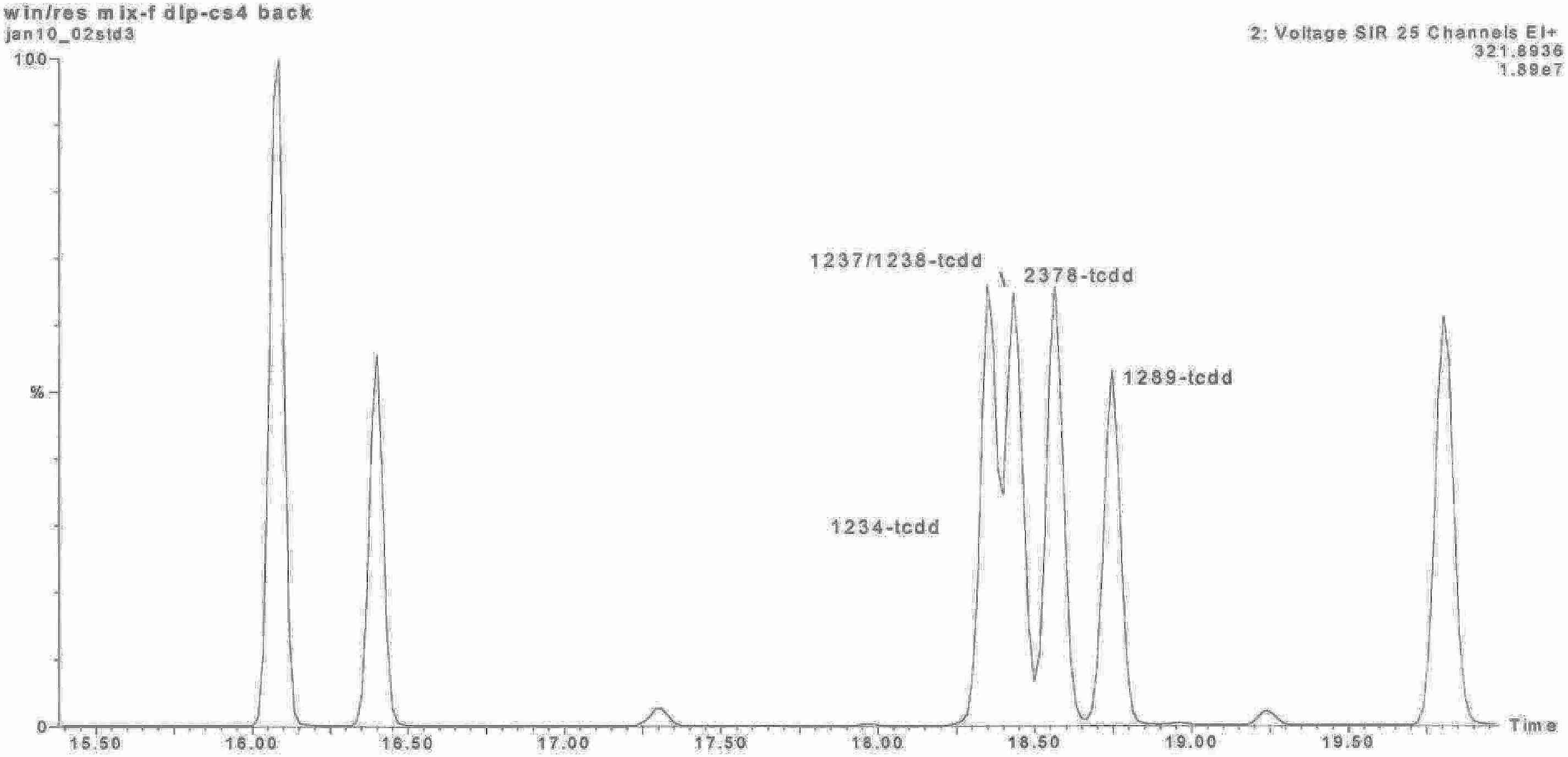
Chromatographic Resolution



Ministry of the Environment

ONTARIO

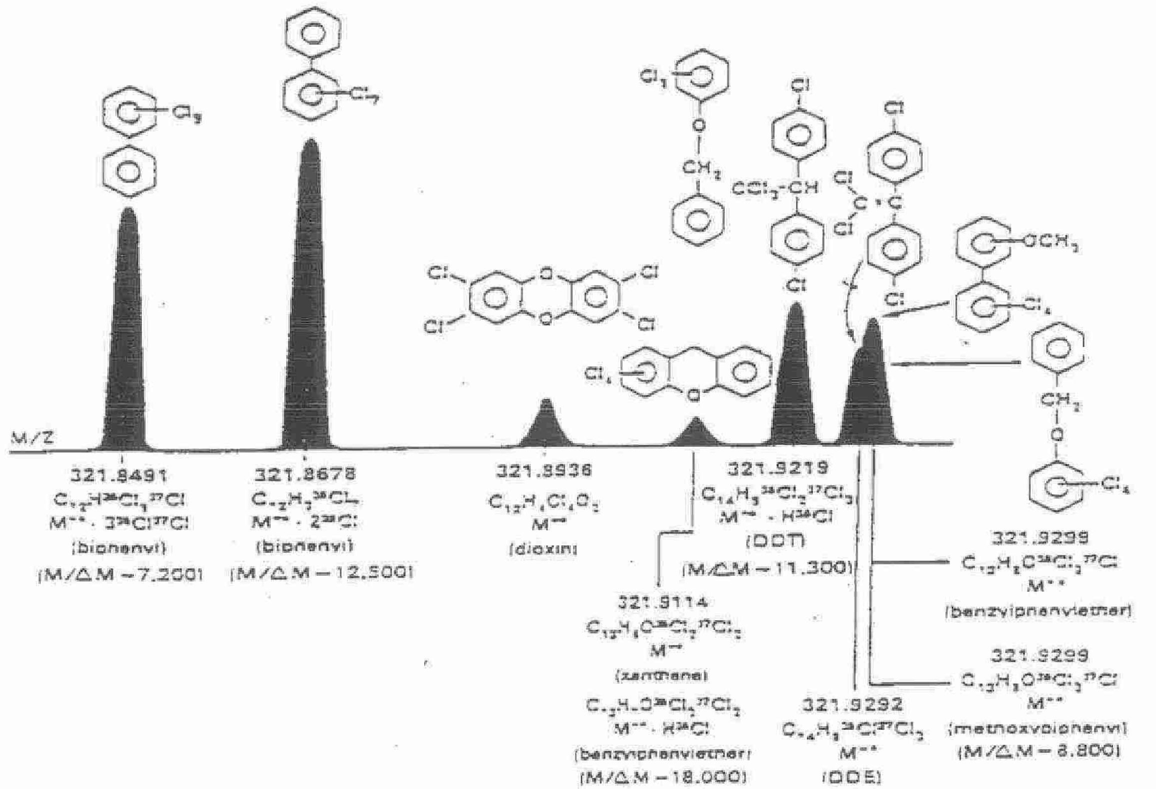
Quality Assurance / Quality Control



Ministry of the Environment

ONTARIO

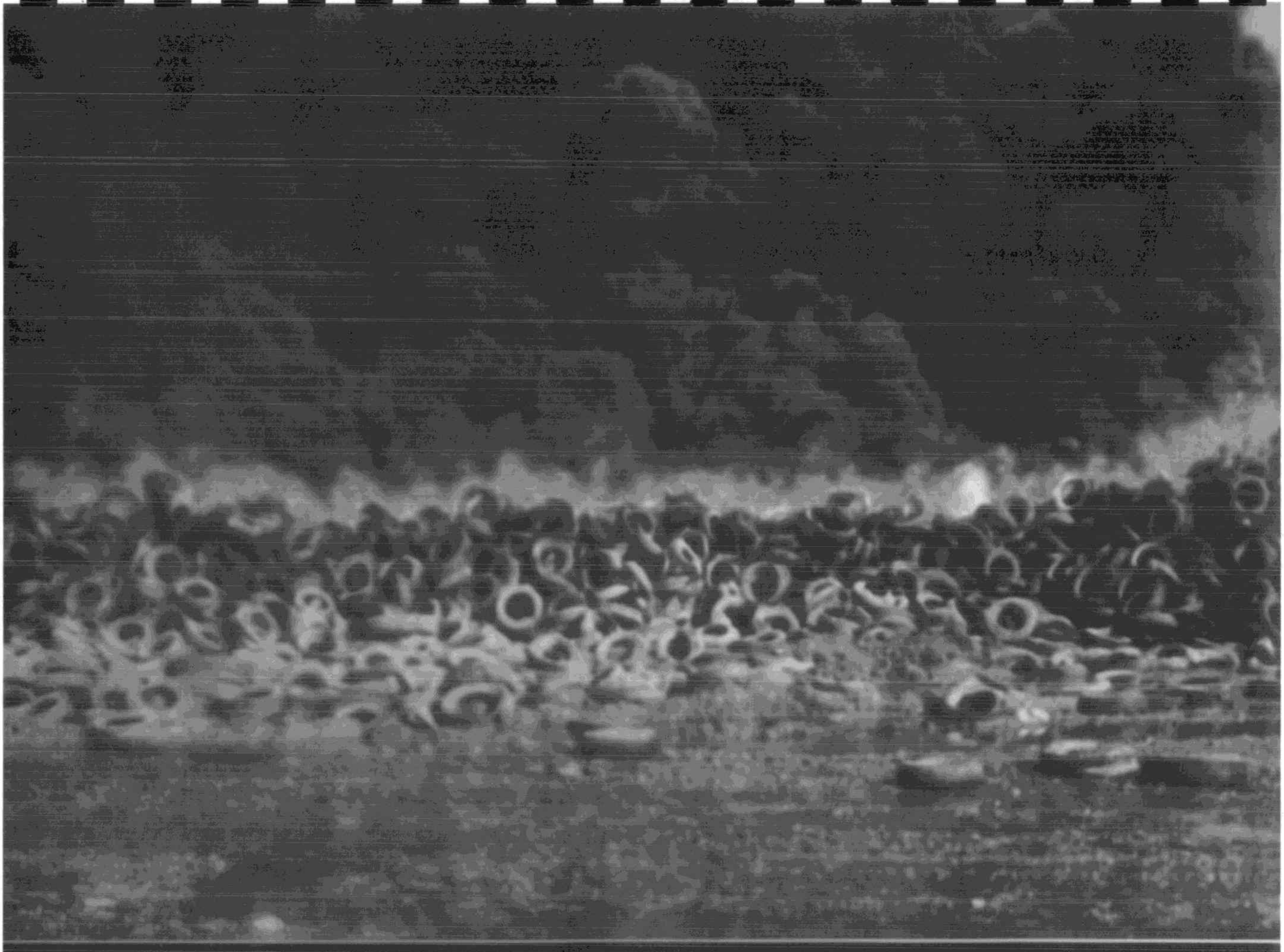
Mass Spectrometric Resolution



Micromass Dioxin Training Manual

Analysis of Dioxin-like Compounds

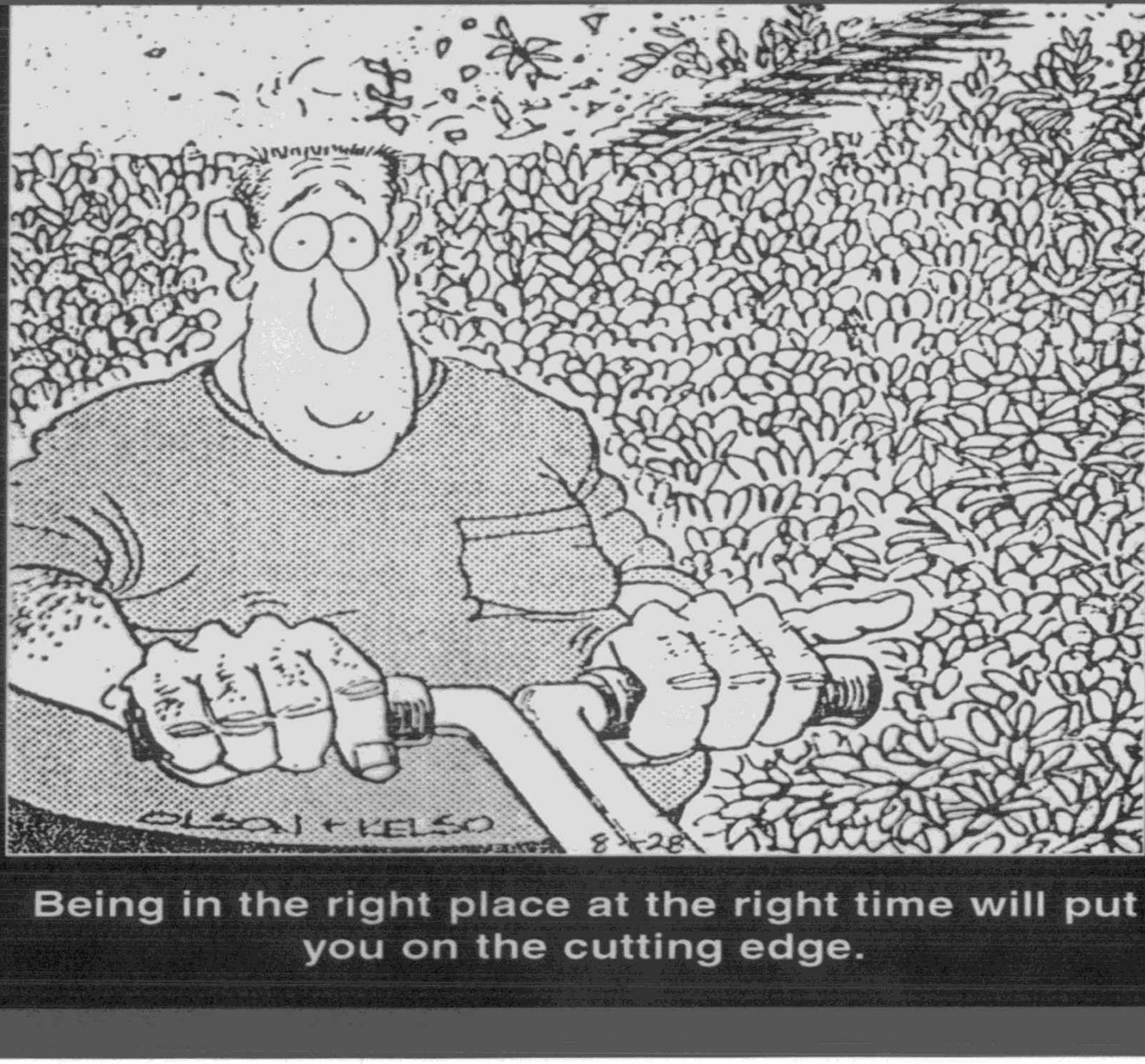
- 8 to 10 days to complete a set of 10 samples for Dioxins and DLPCBs
- 2 sample fractions- Dioxin/Furans & coplanar PCBs
  - Mono-ortho PCBs
- Analytical run times of ~ 45 min / sample with QC samples 10 samples take about 16 to 18 hours to run (for each fraction)



Residents gather to watch smoke and flames billow from the Plastimet plant on Wednesday night.

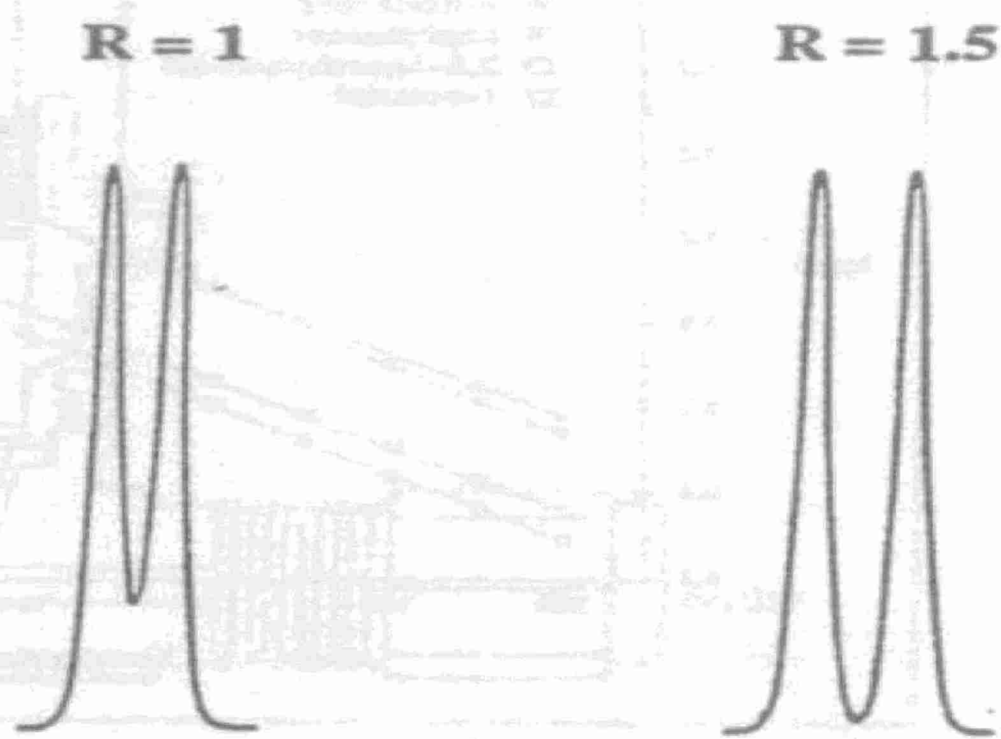
John Rennison, The Spectator

Two-day battle with the monster



Being in the right place at the right time will put you on the cutting edge.

Chromatographic Resolution



Restek GC Seminar

Why Fast GC?

Resolution

$$R = 1/4 \sqrt{L / h} \times (\alpha-1 / \alpha) \times (k / k+1)$$

Efficiency (width of peaks)

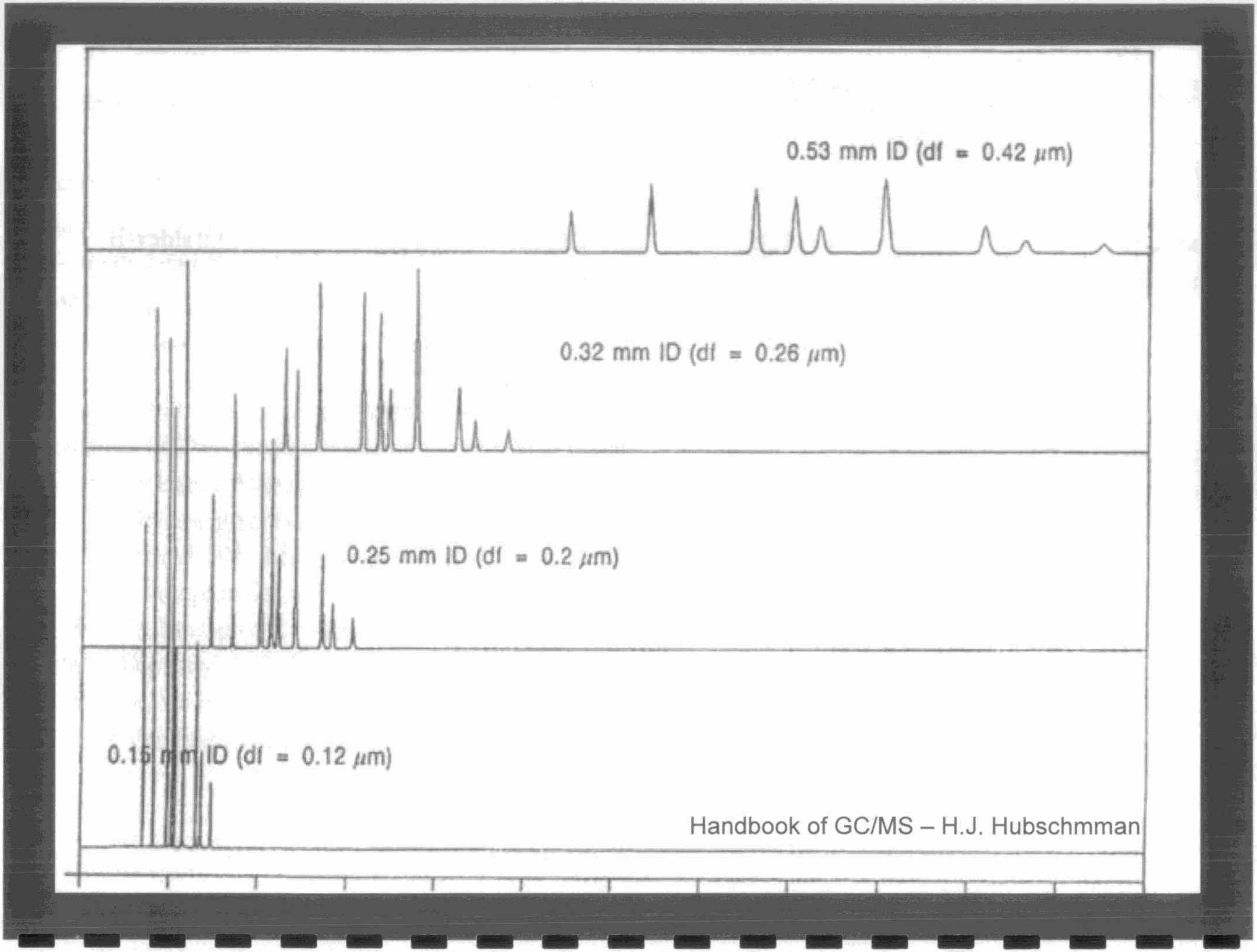
$$N = L / h \quad (h = HEPT) = (t_R / W)^2$$

Selectivity (spacing of peaks)

$$\alpha = k_2 / k_1$$

Capacity Factor

$$k = t_R - t_0 / t_0$$


0.53 mm ID (df = 0.42 μm)
0.32 mm ID (df = 0.26 μm)
0.25 mm ID (df = 0.2 μm)
0.15 mm ID (df = 0.12 μm)
Handbook of GC/MS – H.J. Hubschmman

What is Fast GC?

- GC Ramp Rates >50ºC/min.
- GC column diameters <0.18 mm
- Stationary phase <0.18 µm
- GC column head pressures >60 psi
- Phase Ratio: $\beta = r / d_f$

r = column diameter

d_f = film thickness

Why Use Fast GC?

- Decrease analysis times by up to 80%
- Increase instrument throughput by 4x
- Faster emergency response
- Reduce analysis costs
- Reduced sample sizes & resources
- Same Phase Ratio (film thickness/column i.d.) means same chromatography

Comparison of GC Columns

Column Length (m)	10	30	20	60	40
i.d. (mm)	0.1	0.25	0.1	0.25	0.18
Film Thickness (μm)	0.1	0.25	0.1	0.25	0.18
Theoretical Plates/m	8,600	3,300	8,600	3,300	5,300
Total Theor. Plates	86,600	99,000	172,000	198,000	212,000
Rel. Col. Efficiency	0.93	1	1.32	1.41	1.46

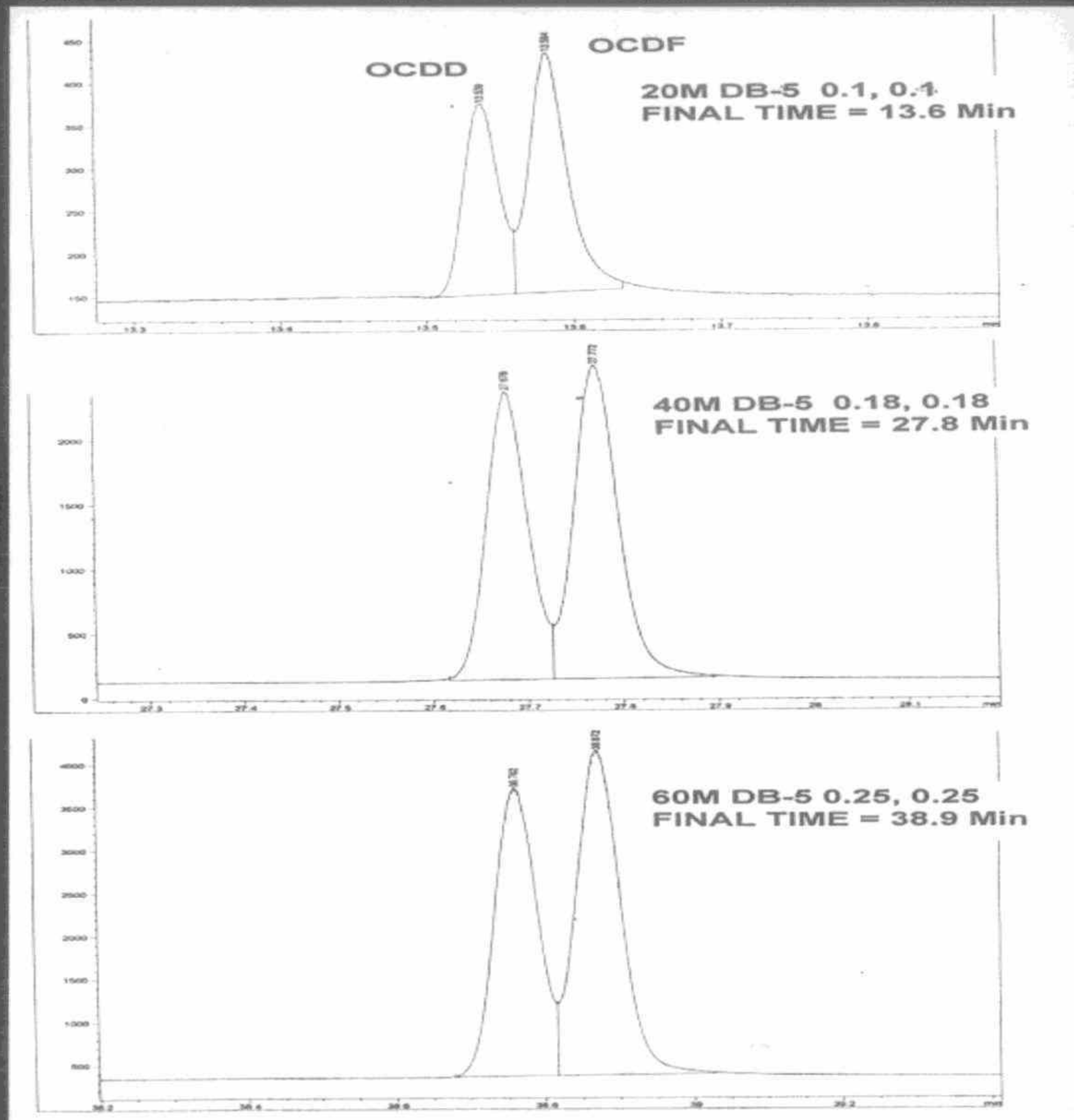
How Can I Do Fast GC?

- TRANSLATE method to Fast GC column using Agilent Technologies Method Translation Software
- OPTIMIZE chromatography using Restek Pro EZ GC
- Use appropriate equipment/GC conditions [GC ramp rates >50° C/min.; 60 psi head pressure]

Column Comparison: Dioxin

Column Length (m)	20	60	40
i.d. (mm)	0.1	0.25	0.18
Film Thickness (μm)	0.1	0.25	0.18
Theoretical Plates/m	8,600	3,300	5,300
Total Plates	172,000	198,000	212,000
Effective Plates (TCDD)	176,000	230,000	285,000
Relative Efficiency	0.93	1	1.03
Relative Anal. Time	0.33	1	0.55

GC Column Comparison Dioxin Analysis: Fast Mode

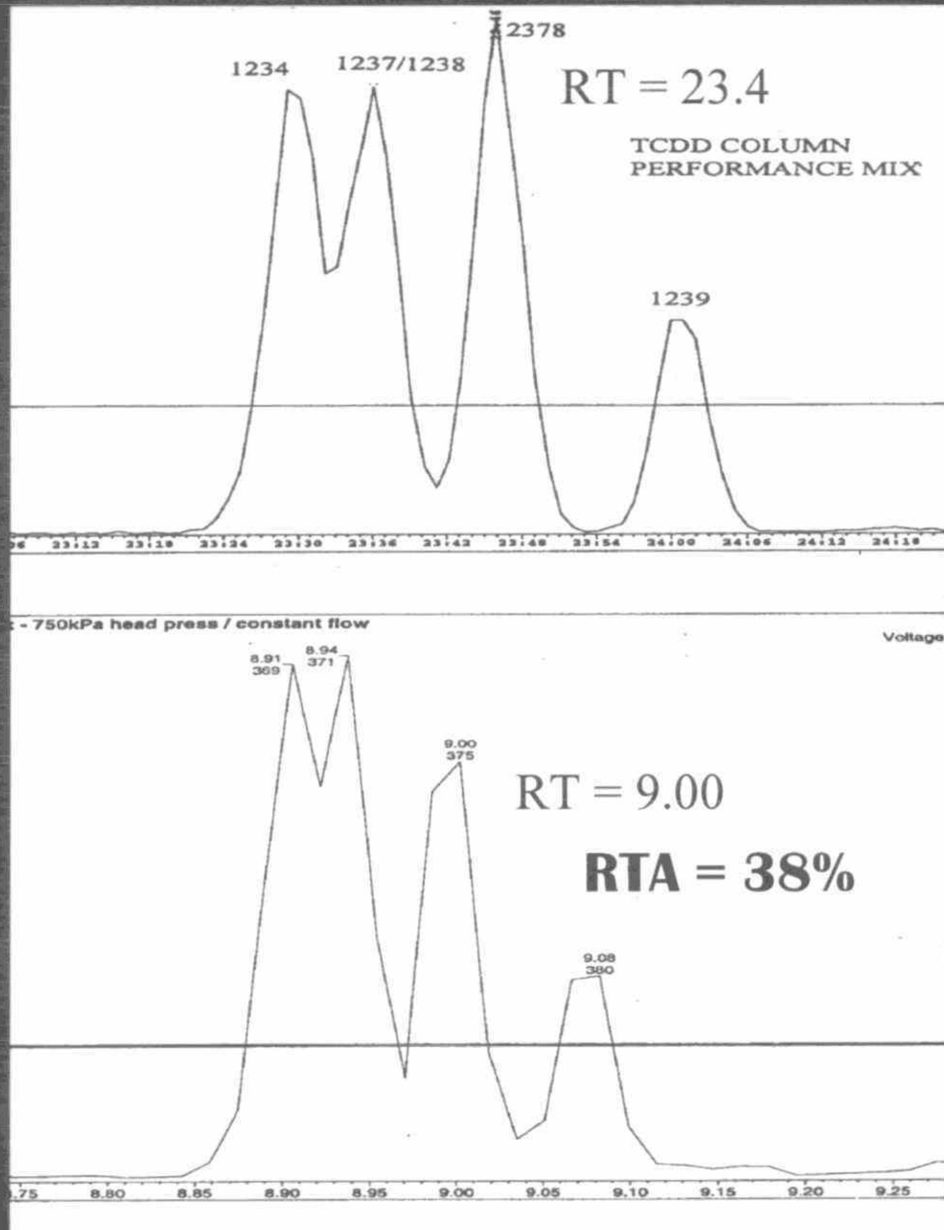


60m - 0.25, 0.25
5% phenyl

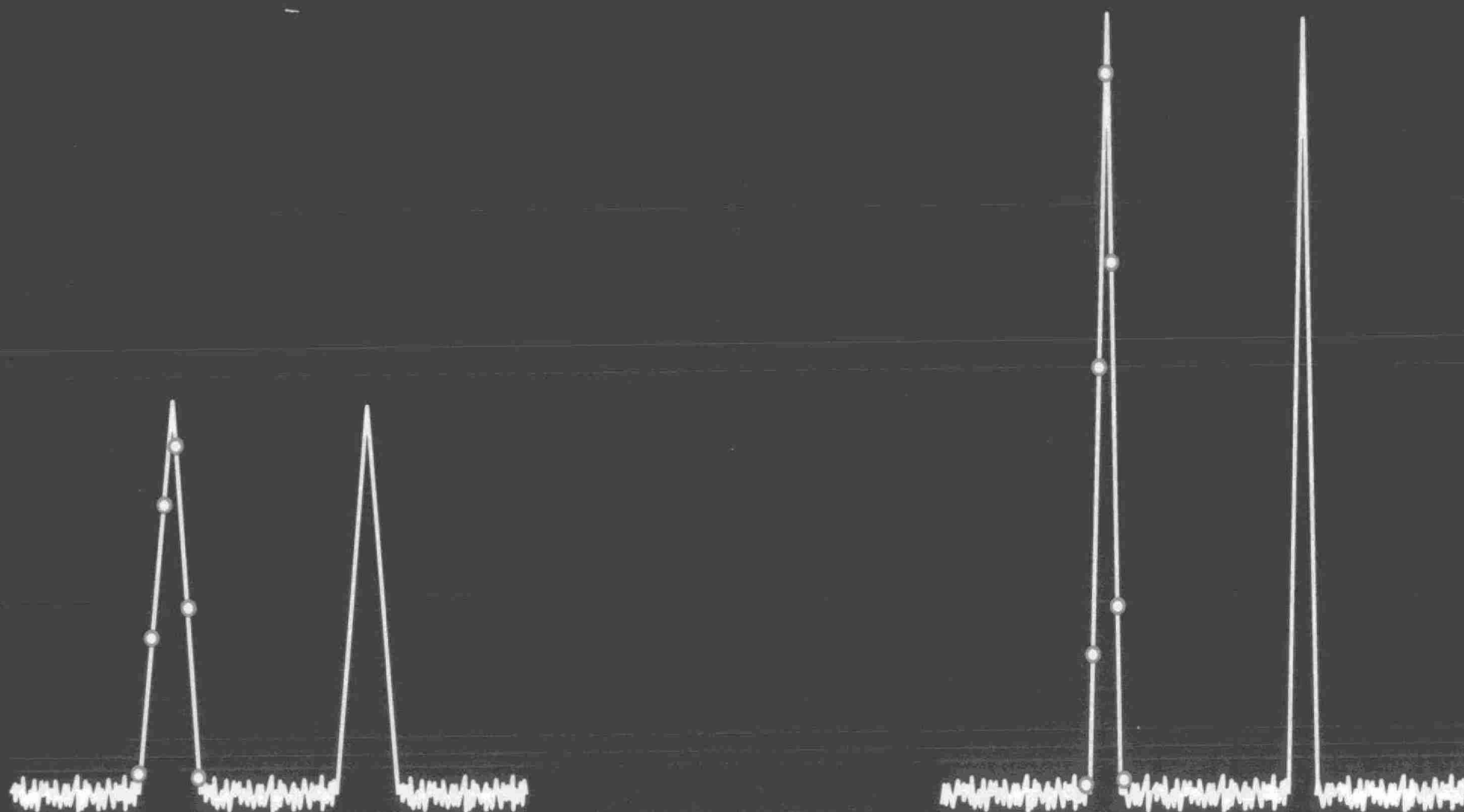
25% Valley

20m - 0.1, 0.1
5% phenyl

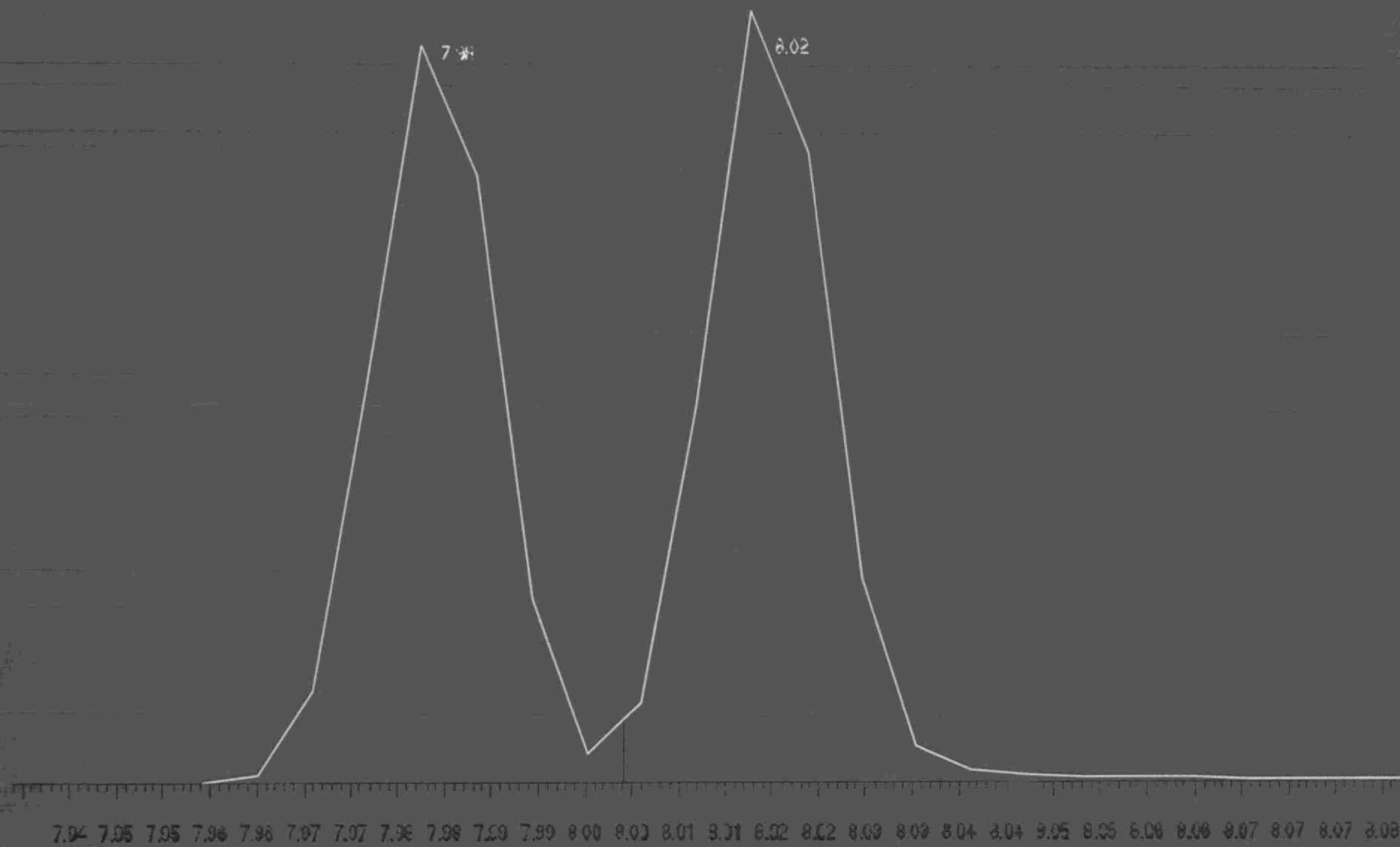
25% Valley



Minimum Data Points / Peak



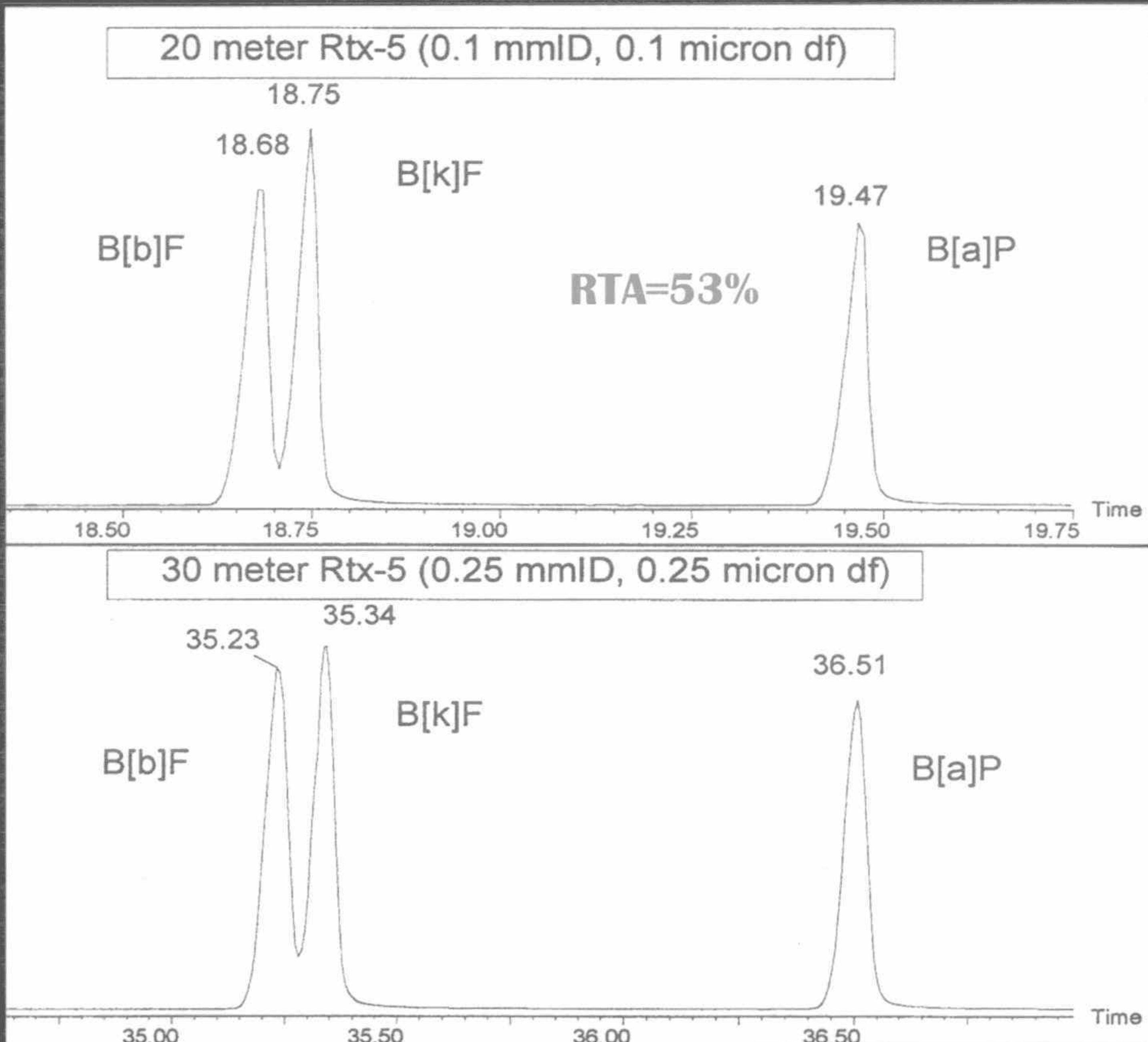
Minimum Data Points / Peak



Column Comparison: PAH

Column Length (m)	10	30	20
i.d. (mm)	0.1	0.25	0.1
Film Thickness (μm)	0.1	0.25	0.1
Theoretical Plates/m	8,600	3,300	8,600
Total Plates	86,000	99,000	172,000
Relative Efficiency	0.93	1	1.32
Relative Anal. Time	0.38	1	0.53

PAH Critical Pair



PAH Results Comparison

PAH	30 m 5% Phenyl (ng/g)	10 m 5% Phenyl (ng/g)	Relative Amount (30 m/10 m)
Phenanthrene	840	830	1.01
Anthracene	200	190	1.05
Fluoranthene	1000	1000	1
Benzo[b]fluoranthene	280	310	0.90
Benzo[k]fluoranthene	280	280	1
Benzo[a]pyrene	280	280	1
Dibenzo[g,h,i]perylene	160	180	0.89
Total Priority 16 PAH	**4920**	**4990**	**0.986**
Total Analysis Time	**40 min.**	**14 min.**	

Critical Issues for Fast GC

- Proper GC liner - 1 mm or 2 mm
- Syringe capable of sub-μL injections [0.2 μL to 0.5 μL]
- Fast detector: need min. 7 scans/GC peak [may need to reduce #ions in scan window]
- Small GC sample tray can limit throughput

Reductions in Analysis Times Using Microbore Columns

	60 m (.25/.25)	40 m (.18/.18)	30 m (.25/.25)	20 m (.10/.10)	10 m (.10/.10)
Dioxins	[50]	28 (44%)		14 (72%)	
PCB Congeners	[90]			18 (80%)	
PAH			[40]	22 (55%)	14 (65%)
OC Pesticides			[55]	12* (78%)	

Fast GC Summary -- I

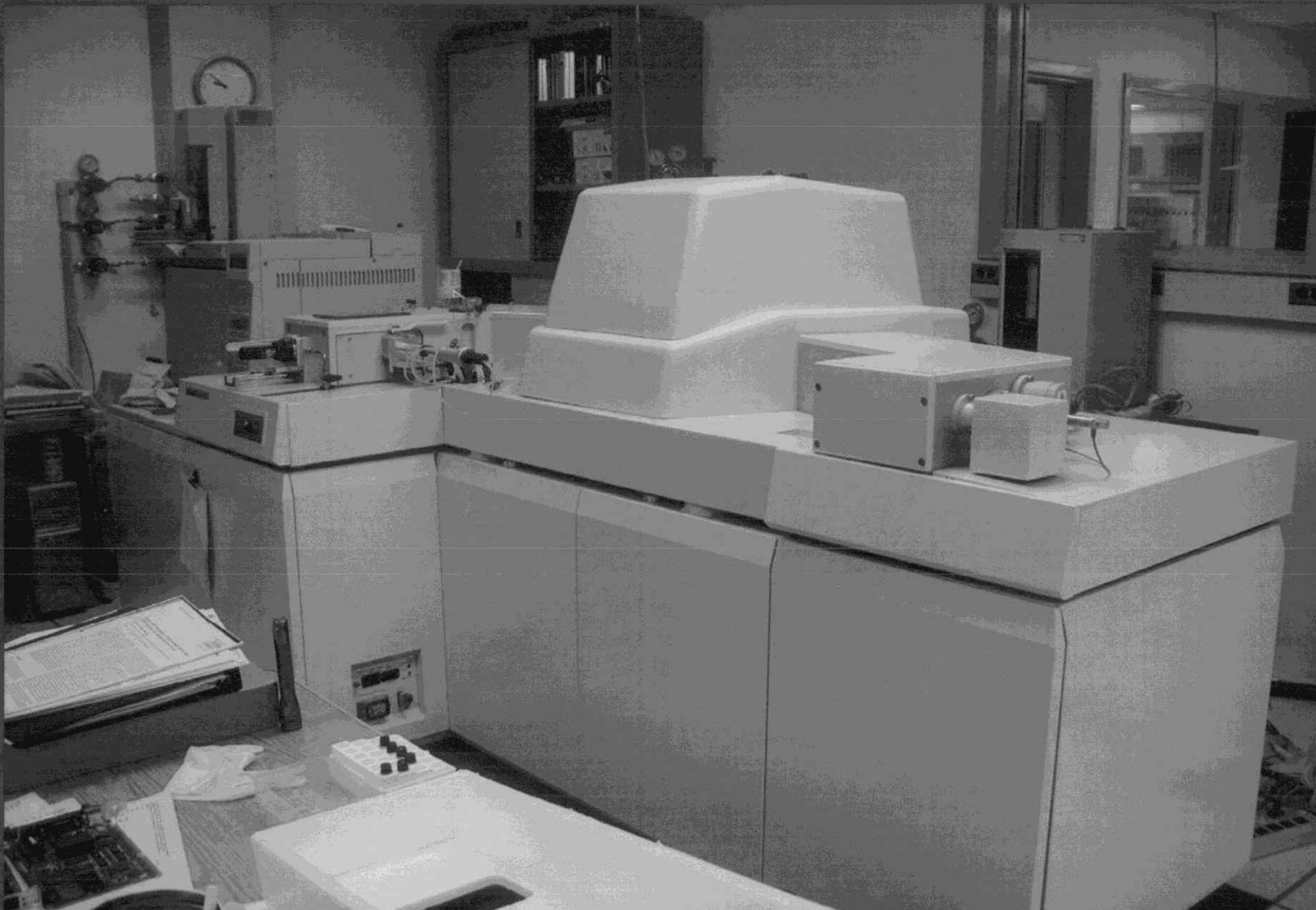
- Fast GC techniques can be applied to many compound types using various GC phases
- GC times 50% to 80% less by converting methods to 10 m and 20 m columns
- Can increase instrument throughput >4x
- Analysis times can be reduced further by use of analyte-specific columns

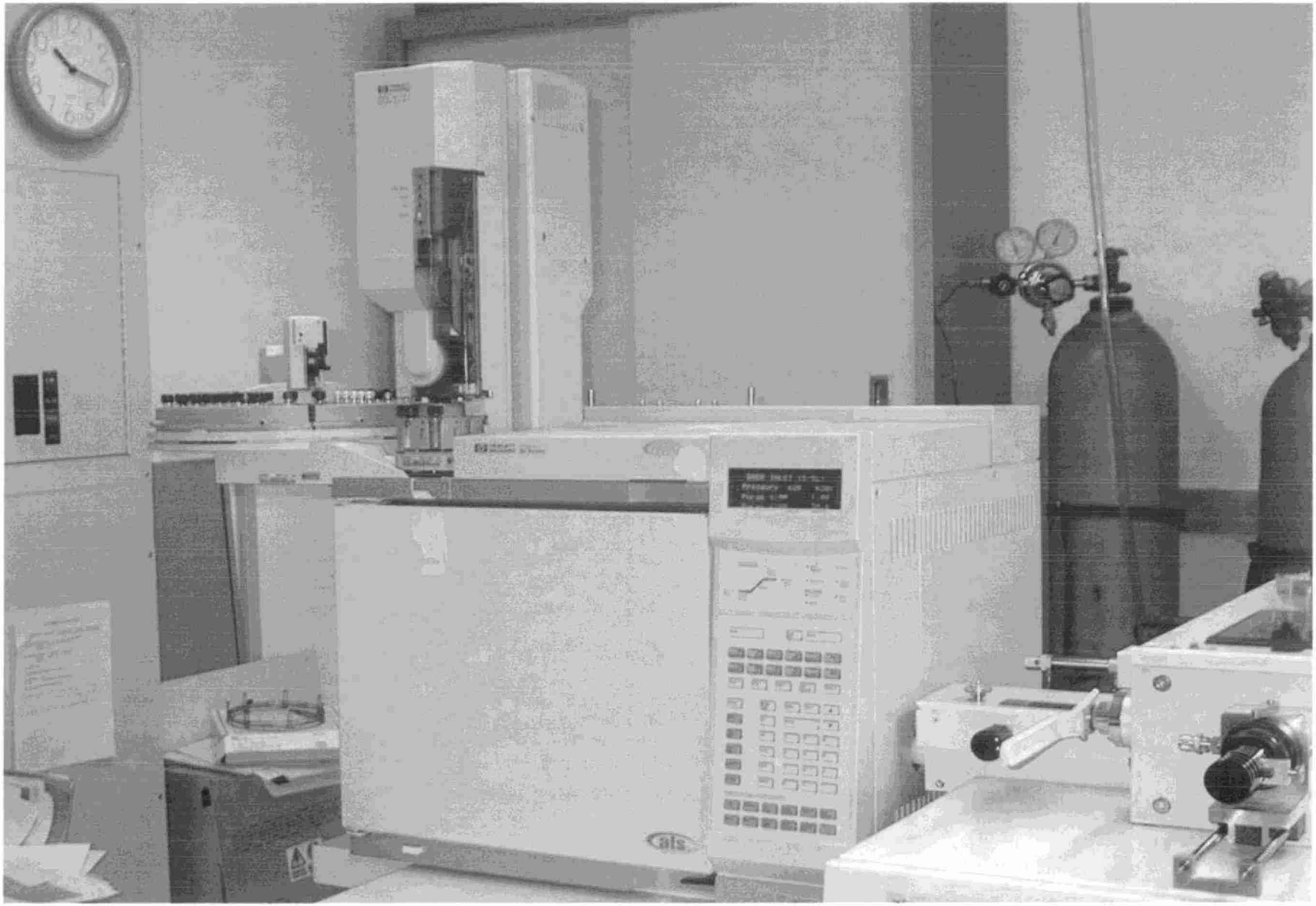
Fast Summary -- II

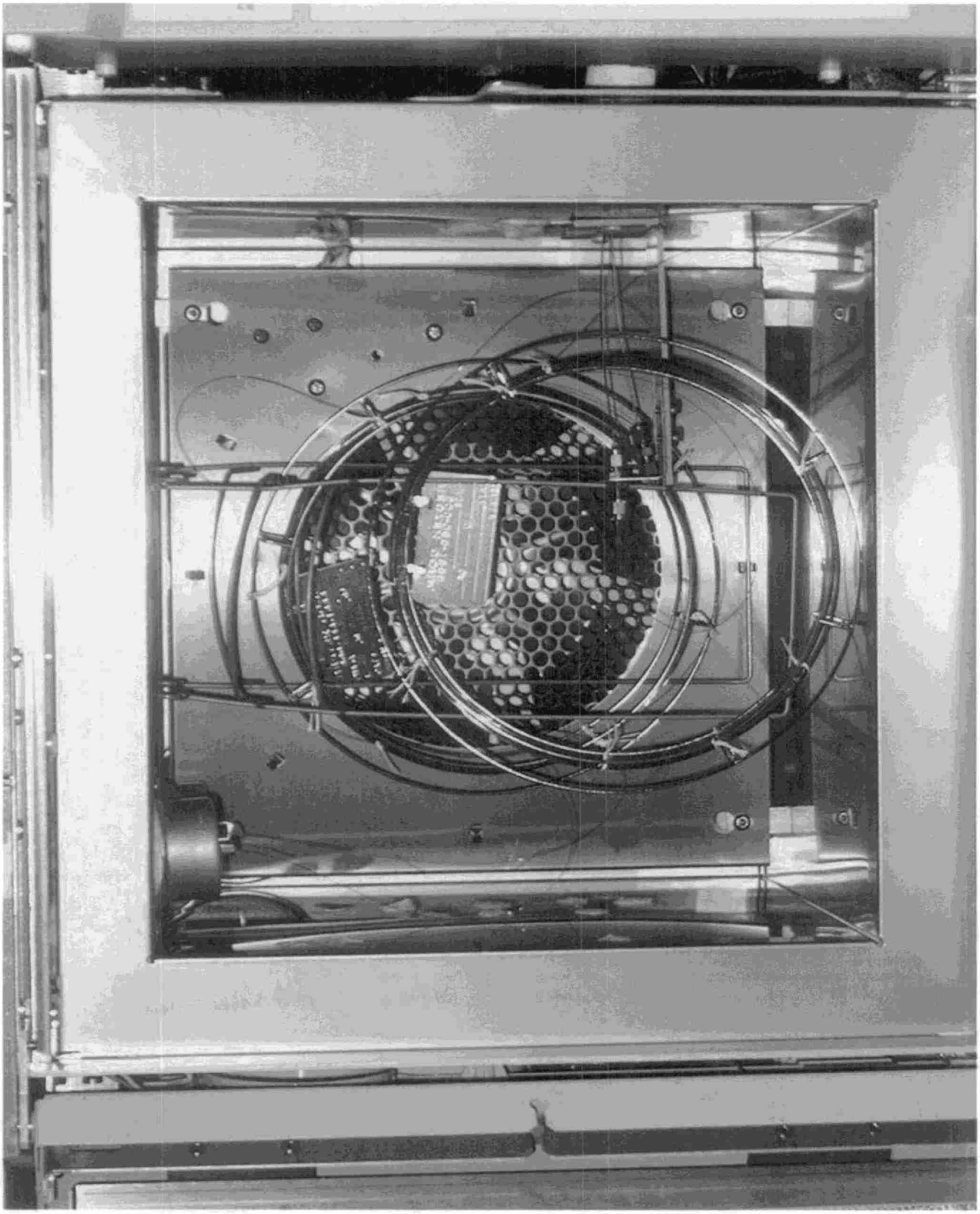
- H_2 reduces analysis times up to 30% over He
- Simple GC optimization (head pressure, ramp rates) can lower analysis times 50% for older GCs not capable of Fast GC operation
- GC-MS Data quality not affected by Fast GC operation unless GC-MS scan rates too slow [minimum 7 scans per GC peak required]
- Regulatory agencies slow to adopt

Activated Carbon

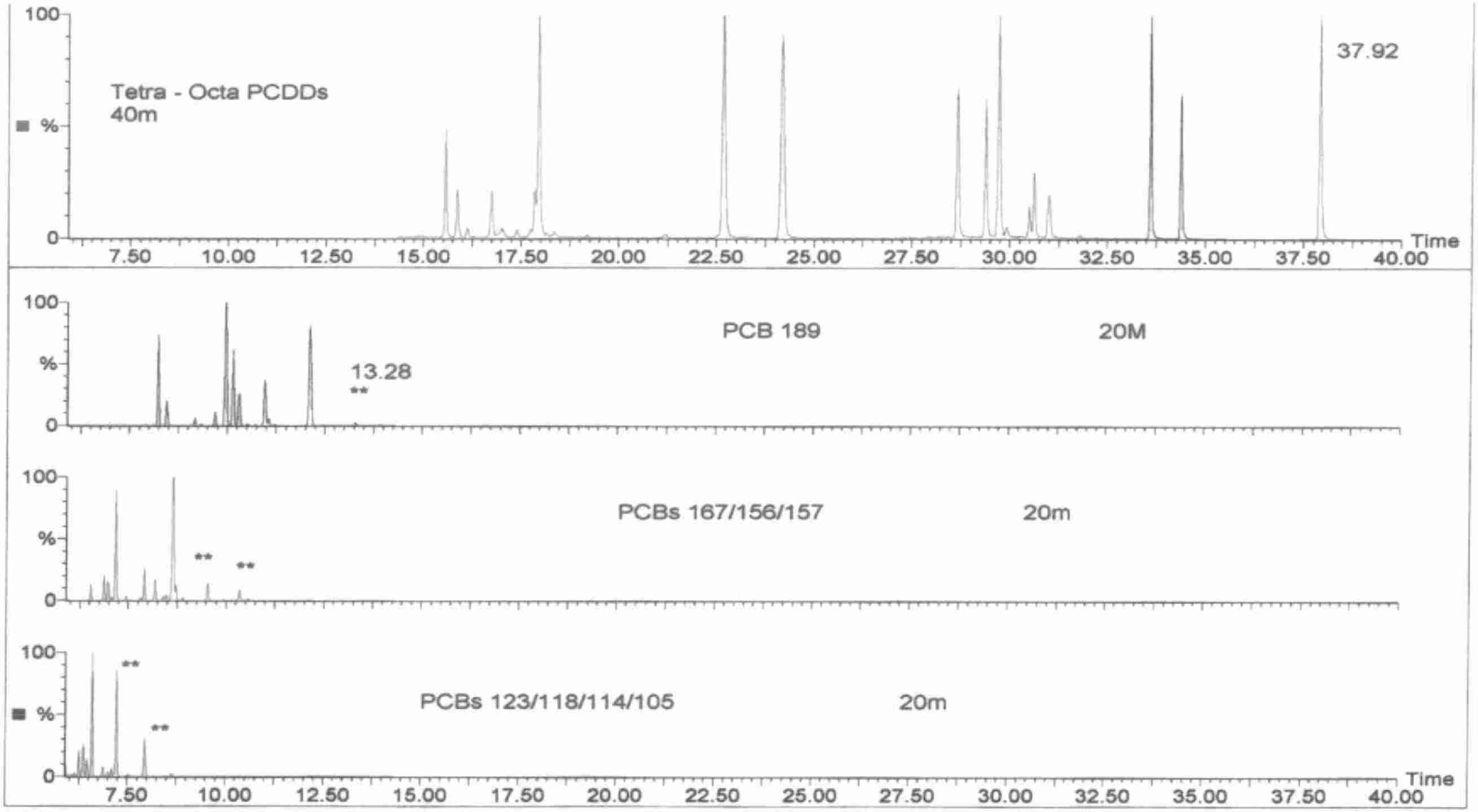
- Planar dioxin-like compounds retained
  (Dioxins, furans, co-planar PCBs, non-ortho CDEs, PCNs)
- Non planar compounds not retained (ortho substituted PCBs, CDEs, BDEs
- Interfering compounds in separate fractions – Furans vs CDEs
  PCB 77 vs PCB 110







PCDDs & Mono-ortho DLPCBs



Dual Column Advantages

- Mono-ortho PCBs elute from 20m column well before PCDD/Fs elute from 40m column
- DPEs, and interfering PCBs in mono-ortho DLPCB sample fraction
- Avoid potential interferences:
  1. Furan formation in ion source from DPEs
  2. Reduces co-elution of higher chlorinated PCBs with Coplanars (i.e. PCB 110 with PCB 77)

Dual column GC conditions

0.5 µl injection	**40m, 0.18mm x 0.20 µm FRONT**	**20m, 0.1mm x 0.1µm BACK**
Head Pressure	**380kPa**	**610kPa**
Initial Temp.	130 °C - hold 1min	
1st Ramp	52 °C/min → 200 °C, 0 hold	
2nd Ramp	2.9 °C/min → 235 °C, hold 10 min.	
3rd Ramp	6.9 °C /min → 300 °C, hold 3.5 min.	
Total Run Time	37.8 min	

Dioxin/Furan Results Comparison

NIST 1944 MARINE SED.	Single (pg/g)	Dual (pg/g)	Ref. Value (pg/g)
2378-TCDD	136	128	133 ±9
12378-PCDD	19	22	19 ±2
123478-HxCDD	29	27	26 ±3
23478-PCDF	48	45	45 ±4
OCDF	1300	1200	1100 ±100

DLPCB Results Comparison

NIST 1944 MARINE SED.	Single (pg/g)	Dual (pg/g)	Ref. Value (pg/g)
PCB 105	24,000	26,000	24,500 ±1100
PCB 118	54,500	62,600	58,000 ±4300
PCB 156	5900	6150	6520 ±660
PCB 126	270	262	N/A
PCB 169	16	14	N/A

Dual Column Timesavings

- Dual column analysis time for Dioxins/Furans & DLPCBs reduced from

90min to 38 min

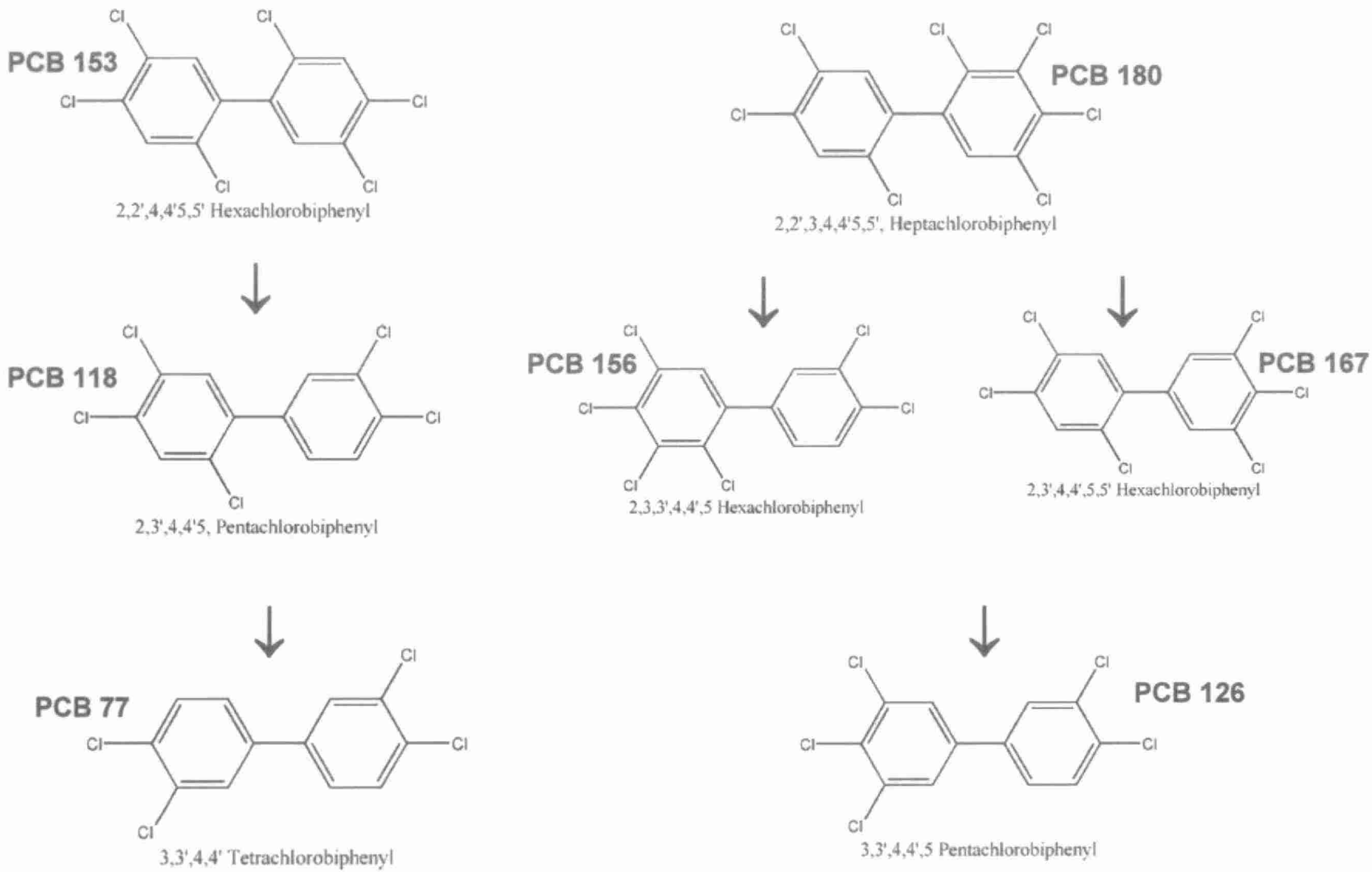
(2 x 45min, 2 separate days)

Greater than 50% Timesavings!

Dioxin-like PCBs

- A few congeners with very high Dioxin-like toxicity (PCB 126)
- Physical, chemical and biological properties more closely related to dioxins
- Can be formed by dechlorination of other more highly chlorinated PCBs
- Relative amounts of DLPCBs increasing

Main Aroclor congeners: 180, 153, 138, 118, 52, 28



Barr et al, Organohalogen Compounds 33, 199, (1997)

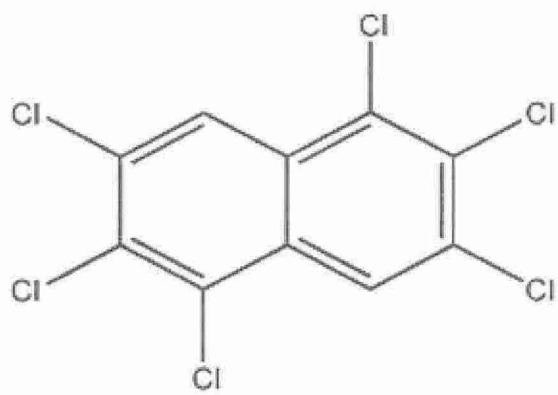
DLPCB Results Comparison - Fish

Lake	PCB TEQ / Dioxin TEQ	n
Superior	4.4	6
Huron	12.9	7
Erie	6.8	1
Ontario	5.4	15
Niagara River	3.8	6

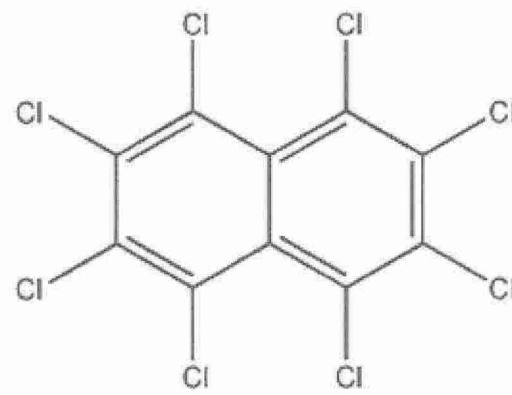
Polychlorinated Naphthalenes

- High Dioxin-like toxicity - HexaCNs
- Planar structure
- Used as protective coatings, dielectric fluids, flame retardants and fungicides
- Use restricted in 1920s due to death of factory workers
- Amount manufactured 10% of PCB total

Polychlorinated Naphthalenes



1,2,3,5,6,7 Hexachloronaphthalene



Octachloronaphthalene

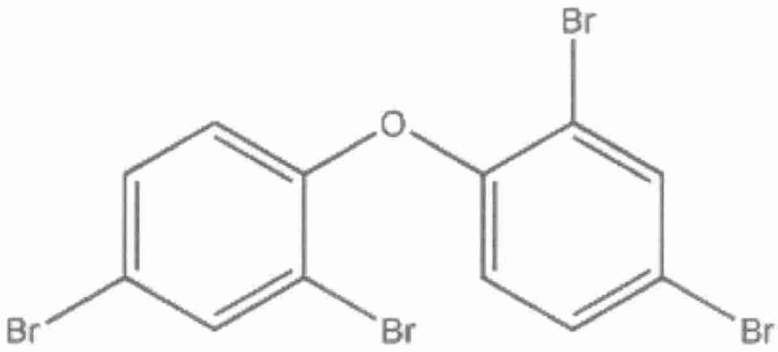
Polychlorinated Naphthalenes

Station No. Detroit River	1160	803	1169	1168	**1161**	**804**	**1159**	1156	1157
Total PCNs (ng/g)	3.5	1.2	8.2	9.2	**7400**	**8.0**	**8200**	6.0	300
PCN TEQs (pg/g)	2.4	1.3	2.0	3.3	**3300**	**4.4**	**2900**	3.7	73
Total Dioxin/PCB TEQs	2.3	3.3	25	18	**310**	**110**	**5.7**	38	8.4
Dioxin TEQs (pg/g)	1.0	2.2	14	11	**280**	**73**	**4.4**	29	5.8
PCB TEQs (pg/g	1.3	1.1	11	6.8	**22**	**35**	**1.3**	9.6	2.6

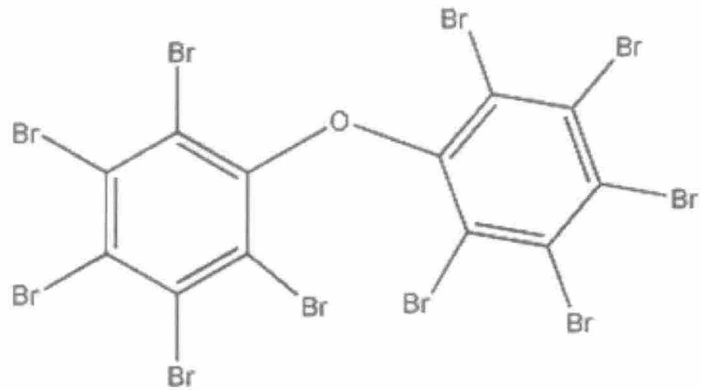
Brominated Diphenylethers

- In USA – 3 million fires resulting in 29000 injuries and 4500 deaths annually
- Second largest additive to plastic materials
- Make up to 30% by weight in foam cushions and plastics
- Health effects include: Thyroid effects, Neurobehavioral effects and cancer

Brominated Diphenylethers



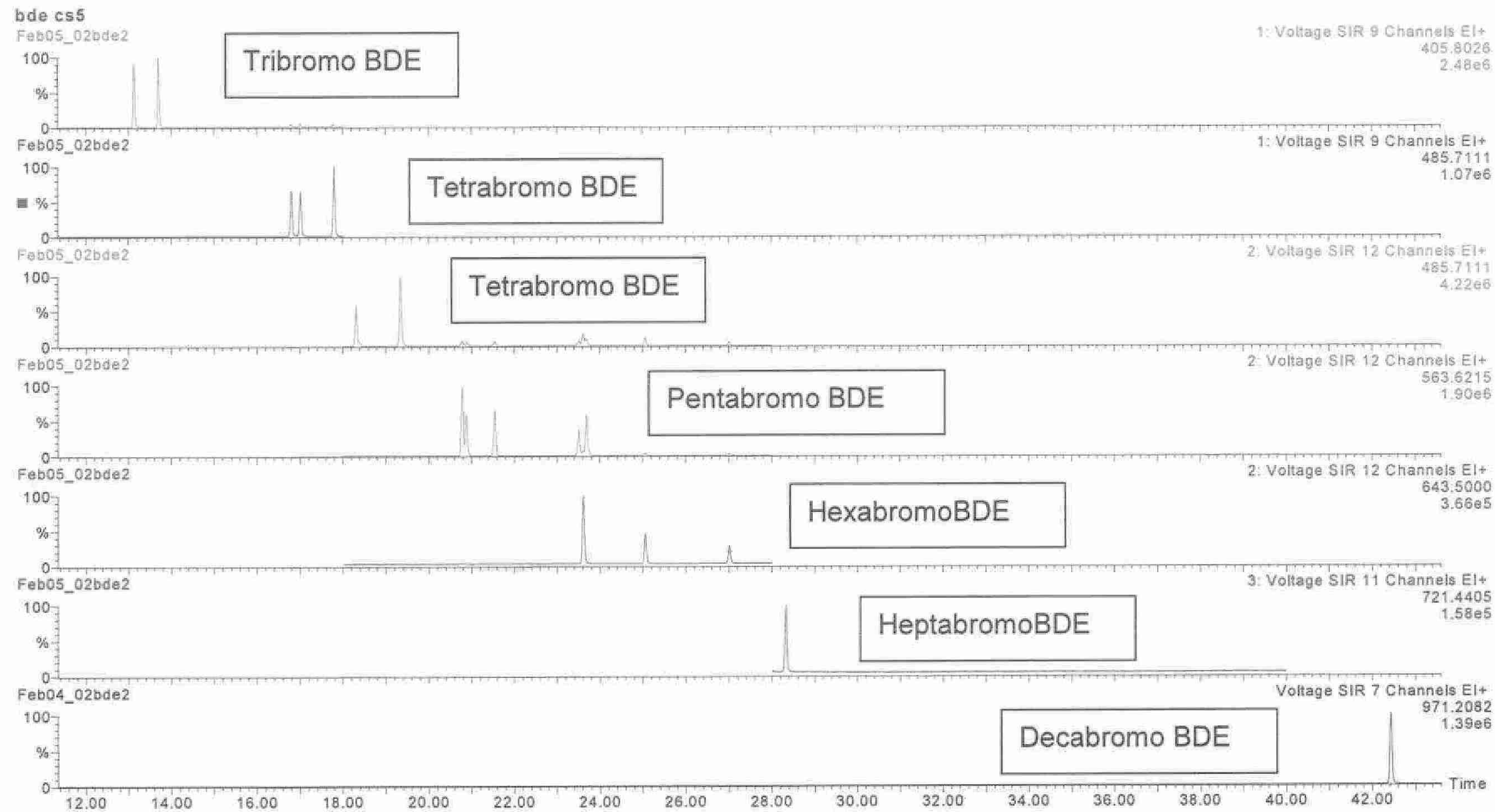
2,2',4,4' Tetrabromodiphenylether



Decabromodiphenylether

Mixture	Use	Congener Composition	Usage (tons - 1999)
1)Penta	Polyurathane foam (furniture)	BDE – 47,99,100,153,154	8500
2) Octa	Rigid Plastic(electronics)	hepta and octa	3825
3) Deca	Rigid Plastic, resins coatings & insulators	BDE – 209	54800

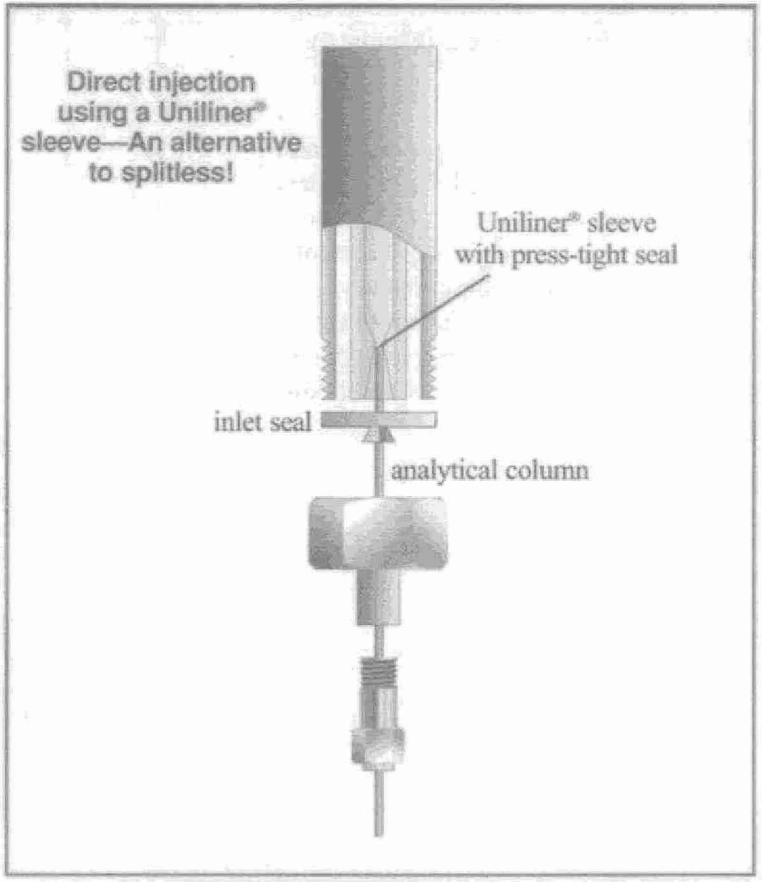
Brominated Diphenylethers



Ministry of the Environment

ONTARIO

Brominated Dyphenylethers



Restek Uniliner Brochure

- Wide range in physical properties (mass, melting point, boiling point)

<u>Mass Range</u> – 230 (mono)
972 (deca)

<u>Deca – BDE</u>

Melting point – 300 °C

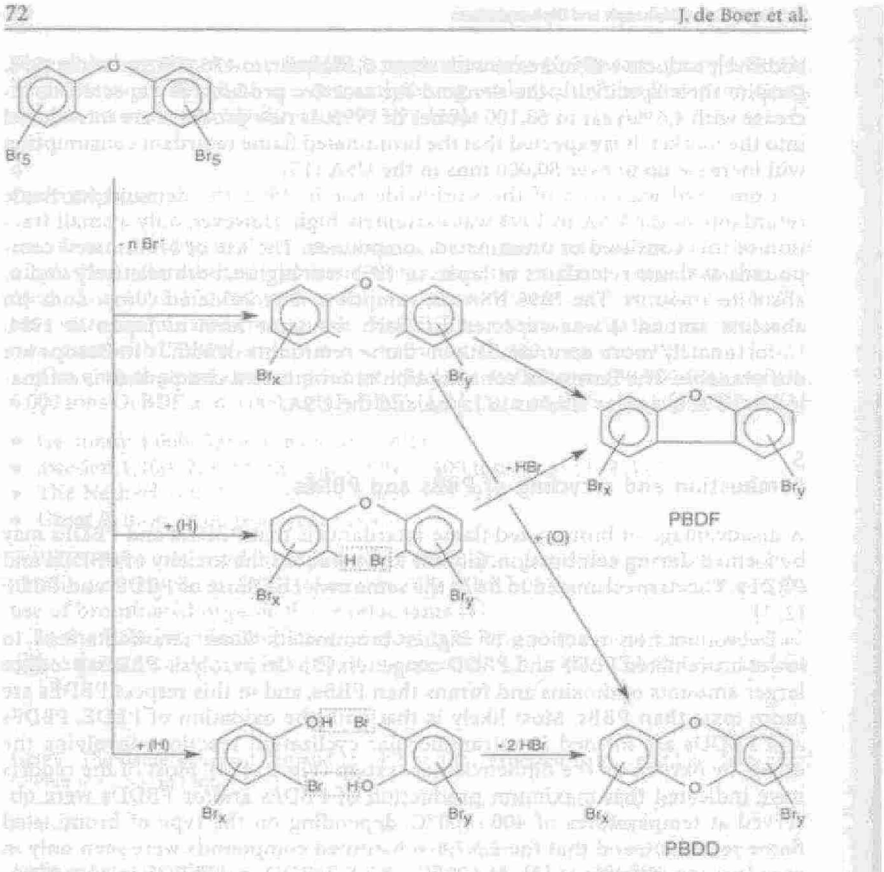
Decomposition - 420 °C

Use high temp carborane column

Future Work

- Compare levels of DLPCBs in environmental samples (seds, water, biota) with Aroclor patterns and determine if weathering results in TEQ increase
- Determine PCN levels in Great Lakes sediments and fish and compare with DLPCBs
- Determine BDE degradation processes

Brominated Diphenylethers
Furture Work



- How do DBE's degrade in foams, plastics and other materials?

- Do the Bromines exchange with Chlorine and form Br / Cl diphenylethers, furans and dioxins?

New Types of Persistent Halogenated Compounds – J. Paasivirta -2000

Acknowledgements

MOE:

K.A. MacPherson T.M. Kolic R. Brunato

T. Chen M.A. Bogard A.R. Boden

G. Ladwig V. Khurana

Environment Canada:

Chris Marvin

Restek:

Frank Dorman

Ministry of the Environment

ONTARIO

	DATE DUE		